Vol. 31 No. 4

UNIVERSITY OF MICHIGAN
DENTISTRY LIBRARY

Journal of DENTAL RESEARCH

Founded by William J. Gies in 1919

OFFICIAL PUBLICATION OF THE INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

Editor

HAMILTON B. G. ROBINSON

AUGUST, 1952

Editor's Viewpoint ---- 453

International Association for Dental Research. Proceedings of the Thirtieth Annual Meeting, Broadmoor Hotel, Colorado Springs, Colo., March 21, 22, and 23, 1952. Compiled by Hamilton B. G. Robinson. (Ohio State University) -- 455

I. President's Address. Maynard K. Hine. (Indiana University) ---- 455

II. Index of participants and sequence numerals for corresponding abstracts ---- 458

III-IX. General Sessions: Abstracts 1-95 ---- 459

X. Dental Materials Group: Abstracts 96-104 ---- 496

XI. Papers read by title: Abstracts 105-146 ---- 499

XII. Executive proceedings and register of newly elected officers and members ---- 516

The Effect of the Changing Dentition on Masticatory Function. F. R. Shiere and R. S. Manly. (Tufts College Dental School, Boston, Mass.) ---- 526

Variations in Structural Morphogenesis of the Human Face and Jaws. Initial Stages of Normal and Abnormal Bone Formation (Gross) Exhibited in the Cleared Embyro and Fetus. Samuel Rabkin. (Winter Park, Fla.) ---- 535

(*Contents continued on inside back cover*)

Published for THE INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH
By The C. V. Mosby Company, St. Louis 3, Mo.

Copyright 1952 by The International Association for Dental Research

INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

EDITORS OF THE JOURNAL OF DENTAL RESEARCH

Editor Emeritus—William J. Gies

Publication Committee—J. Frank Hall, Paul E. Boyle, and Hamilton B. G. Robinson.

Editor—Hamilton B. G. Robinson (Ohio State University). *Business Manager*—J. Frank Hall (Indiana University). *Circulation Manager*—Paul E. Boyle (University of Pennsylvania).

Assistants to the Editor—Paul C. Kitchin and J. B. Brown.

Board of Editors—Editor emeritus, members of the Publication Committee, and sectional representatives named in the fifth column below.

THE SECTIONS—THEIR SECRETARIES, AND REPRESENTATIVES IN THE COUNCIL AND IN THE BOARD OF EDITORS (1951)

Division	*Section*	*Secretary*	*Councillor*	*Editor*
North America	Halifax	J. S. Bagnall	J. S. Bagnall	J. S. Bagnall
	Toronto	R. G. Ellis	R. G. Ellis	R. G. Ellis
	Winnipeg	E. Roy Bier	H. J. Merkeley	E. Roy Bier
	Ann Arbor	Philip Jay	Philip Jay	Philip Jay
	Baltimore	M. S. Aisenberg	E. C. Dobbs	J. Ben Robinson
	Birmingham	L. Robinson	J. F. Volker	J. P. Lazansky
	Boston	P. H. Keyes	R. S. Manly	R. O. Greep
	Chicago	B. G. Sarnat	F. J. Orland	G. B. Denton
	Cleveland	S. W. Chase	T. J. Hill	S. W. Chase
	Columbus	H. B. G. Robinson	L. F. Edwards	P. C. Kitchin
	Detroit		P. C. Lowery	
	Houston	I. R. Telford		S. S. Arnim
	Indianapolis	R. W. Phillips	J. F. Hall	M. K. Hine
	Iowa City	A. O. Klaffenbach	A. W. Bryan	G. S. Easton
	Kansas City	Ben W. Warner	H. L. Myers	N. A. Moore
	Louisville	R. E. Myers	D. Y. Burrill	D. Y. Burrill
	Minnesota	D. F. Chernausek	W. D. Armstrong	S. A. Lovestedt
	Nebraska	D. A. Keys	D. A. Keys	R. T. Ireland
	New Orleans	H. W. Peterson	H. W. Peterson	F. W. Wolfe, Sr.
	New York	Isaac Neuwrith	G. Bevelander	F. Krasnow
	Pacific Northwest	N. M. Phatak	H. J. Noyes	B. O. A. Thomas
	Philadelphia	N. B. Williams	P. E. Boyle	S. L. Rosenthal
	Pittsburgh	T. W. Brand	W. F. Swanson	E. G. Meisel
	Richmond	H. D. Coy	A. D. Brashear	O. W. Clough
	Rochester	J. W. Hein	E. R. Costich	
	San Francisco	S. Epstein	W. C. Fleming	Hermann Becks
	St. Louis	E. A. Jasper	W. B. Gurley	E. F. Allen
	Southern Calif.	L. A. Bavetta	B. Z. Rabinowitch	R. L. Rutherford
	Tennessee	J. T. Ginn	O. A. Oliver	
	Washington	J. S. Oartel	E. G. Hampp	D. B. Scott
Not Belonging to a Division	Budapest	Ervin Lörinczy		
	Chengtu	R. G. Agnew	R. G. Agnew	R. G. Agnew
	Denmark	P. O. Pederson		
	Johannesburg	J. Staz		
	London	M. A. Rushton		
	Mexico	F. R. Leycegui	F. R. Leycegui	S. Fastlicht
	Palestine	S. Lewin-Epstein		
	Prague	F. Neumann		
	Austria, Vienna Center	F. Driak		
Dental Materials Group		R. W. Phillips	F. A. Peyton	N. O. Taylor

OFFICERS OF THE ASSOCIATION

President—M. K. Hine, *President-elect*—F. A. Arnold, Jr., *Vice-president*—G. C. Paffenbarger, *Secretary-Treasurer*—E. H. Hatton, *Assistant to the Secretary-Treasurer*—D. Y. Burrill, *Editor*—H. B. G. Robinson, *Trustees*—H. Trendley Dean, P. C. Kitchin, T. J. Hill, H. C. Hodge, L. M. Waugh.

Vol. 31, No. 4, August, 1952. Journal of Dental Research is published bi-monthly by The C. V. Mosby Company, 3207 Washington Blvd., St. Louis 3, Mo. Subscription price: United States, its Possessions, Pan-American Countries, $9.00 per volume; Canada and other foreign countries, $9.50. Single number when available, $1.25. Entered as second-class matter June 24, 1919, at the Post Office at St. Louis, Mo., under the Act of March 3, 1879. Printed in the U. S. A.

INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

Organized: New York City, Dec. 10, 1920. Of those at the meeting of organization, the following remain active members: Adolph Berger, Theodor Blum, G. W. Clapp, H. S. Dunning, W. J. Gies, A. H. Merritt, B. B. Palmer, L. M. Waugh.

Objects (Const., Art. II): . . ." (a) To promote broadly the advancement of active research in all branches of dental science and in related phases of the sciences that contribute directly to the development of dentistry, and which add to the knowledge of the mouth and teeth and of their relations to the body as a whole; . . . (b) to encourage and facilitate coöperative effort and achievement by, and mutual helpfulness among, investigators in all nations in every division of stomatology; (c) to the end, particularly, that dentistry may render cumulatively more perfect service to humanity." (The Association has been using "dentistry" and "stomatology" as synonyms.)

Membership: total (Nov. 10, 1946), 627. *Eligibility (Const., Art. 14, Sec. 1).*—"Any person who has conducted, and published an account of, a meritorious original investigation in dental science, or in any of the sciences contributory to stomatology, shall be eligible to membership in this Association, providing such person conforms to the recognized standards of professional ethics." *Election.*—New members may be elected *only at general meetings of the Association.* Membership in a section is secondary to membership in the Association as a whole. Removal of members from one geographic center to another does not affect their membership in the Association.

Endowment fund of the International Association for Dental Research (as of June 1, 1949): $1,506.06. There have been no expenditures from this fund.

INFORMATION FOR PROSPECTIVE CONTRIBUTORS, SUBSCRIBERS, OR ADVERTISERS

The JOURNAL OF DENTAL RESEARCH was dedicated by its founder to the dissemination of knowledge pertaining to the teeth and mouth, and to their relations to the body as a whole. It publishes original investigations in these and related fields. Articles are accepted only on condition that they are contributed solely to this JOURNAL. Articles appearing in other domestic or foreign periodicals will not be reprinted in the JOURNAL except in extraordinary cases. The JOURNAL will reproduce a limited number of half-tones or zinc etchings without cost to the author. References to the literature must conform to the following style:

1. (To a periodical)—Doe, J. A.: Dental Disease, *J. D. Res.* **16**: 42, 1937.
2. (To a book)—Doe, J. A.: The Dental Practice, ed. 2, Chicago, 1930, Smith, ch. 2, p. 114.

See McBride, *J. D. Res.* **16**: 537, 1937.

Titles should be given for all references. Papers presented at society meetings should be so footnoted. Reprints, up to 500 copies, will be supplied at prices quoted by the publisher after the JOURNAL is in the mail, and must be ordered from the publisher. Additional reprints may be purchased only at the discretion of the Board of Editors.

Subscription price for the volume (issued bimonthly beginning with February) is $9.00; Foreign, $9.50. Single copies $1.85. The price of volumes and single numbers prior to 1948 will be quoted upon request to the office of the Business Manager, 1121 W. Michigan St., Indianapolis, Ind.

Matters pertaining to subscriptions, change of address, and other business matters should be addressed to The C. V. Mosby Company, Publishers, 3207 Washington Blvd., St. Louis 3, Mo.

Address all communications relating to editorial matters to the JOURNAL OF DENTAL RESEARCH, Ohio State University, College of Dentistry, Columbus 10, Ohio; advertising or business matters to the JOURNAL OF DENTAL RESEARCH, The C. V. Mosby Company, 3207 Washington Blvd., St. Louis 3, Mo.

EDITOR'S VIEWPOINT

The Proceedings of the International Association for Dental Research (p. 455) include the abstracts of 95 papers read in General Session at Colorado Springs, 9 read before the Materials Group, and 42 published as read by title. These abstracts include a wealth of information. The following are examples of the questions in the minds of dentists that are answered:

Is penicillin-containing dentifrice of value in reducing caries and gingivitis? (Abstracts 5, 6, 38, 41, 136.)

What have clinical tests indicated about the anticaries effect of chlorophyl-containing dentifrices? (Abstract 40.)

Do patients become more proficient in use of artificial dentures? (Abstract 78.)

Are self-curing resins standardized as restorative materials? (Abstract 144.)

At what age does intraoral carcinoma become clinically apparent? (Abstract 146.)

These Proceedings are far more than a report of a scientific society. Their perusal may attract attention to important data, ideas, and procedures that can be used by other investigators and clinicians. These short abstracts permit publication of reports on work in progress and reduce the barriers against rapid dissemination of knowledge.

The masticatory performance of the dentition of children changes as dentition develops. Shiere and Manly (p. 526) observed that eruption of permanent molars did not improve masticatory function immediately but, rather, that efficiency was increased over the three- or four-year post-eruptive period. The lower masticatory performance of children (about one-half that of adults with natural dentition) may be compensated by better gastrointestinal function but it might be questioned whether the lower masticatory efficiency in Class III malocclusion is ever compensated. Correlated nutritional studies of individuals with high and low masticatory performance should give important clues to the value of intact dentitions and are long overdue.

Rabkin (p. 535) collected a group of human fetuses and embryos to use in the study of the morphogenesis of the human face and jaws. He has noted some extremely interesting variations in the development of the fetal faces and jaws before they have been exposed to post-natal trauma and disease. He proposes that individual variations are the results of inherent patterns and that the tendency toward deformities in facial and jaw deformities in civilized populations is especially evident where random matings (with diversity of inheritance) have resulted in diverse physical types. Rabkin also points out that his study throws doubt on the possibility of undernourishment being a cause of alterations in the morphology in the head during prenatal life, with the possible exception of hypercalcemia. This study is a tribute to the work of an individual who has carried out his studies without the benefits afforded members of institutional staffs. His work was begun while a practitioner in dentistry and has been carried on during the author's retirement.

Lisanti and Zander (p. 548) applied heat, in vivo, to the floors of cavities prepared in dogs. They found that much of the heat is dissipated between the cavity floor and the pulp, dentin being a good insulator. Heat ranges, such as those produced in cavity preparation, resulted in inflammatory reactions, including blister formation. The pulps healed after severe injury from temperature elevation. This ability to heal should not be regarded as license to create heat in cavity preparation because: (1) these were healthy pulps, (2) the subjects were dogs which may have greater healing ability and (3) the cavities were filled with zinc-oxide eugenol which has qualities that may aid in repair of injured pulps. This study should, however, increase respect for the healing power of the pulp, a property once given little credit.

Medak, Weinrich, Sicher, Weinmann, and Schour (p. 559) found profound changes in the teeth and supporting structures of developing teeth of rats subjected to single large doses of irradiation. Pulpal and dentinal changes were more profound than the ameloblastic and enamel changes at given dosages. These effects are clinically important in the child with developing teeth and whose jaws may be subjected to heavy doses of radiant energy in an effort to remove soft tissue lesions of neoplastic and other types. The changes described are apparently unrelated to the effects seen secondarily in the teeth of adults receiving heavy irradiation over salivary glands.

Van Huysen and Boyd (p. 575) used color photographs to determine the effectiveness of various dentifrices in cleaning the teeth. Their method offers an approach to studies of the cleansing properties of dentifrices. While it has been generally recognized that accumulations of debris on the teeth are causes of gingivitis, it has not yet been determined that the stain which is removed by toothbrushing, and revealed by the methods used by the authors, is the type of material which causes gingivitis and other periodontal diseases. It will be well to evaluate the importance of this factor as a cause of periodontal disease and gingivitis and to establish the effectiveness of the brush and dentifrice as agents in the prevention of periodontal disease.

Kalnins (p. 582) has investigated the origin of enamel drops and cementicles in the continuously growing teeth of rodents. He found that enamel drops were formed by active ameloblasts which had been detached from the surface of growing enamel matrix. The presence of free enamel drops led him to the conclusion that the presence of dentin is necessary only for the beginning of amelogenesis. This is in accord with a similar finding of Fleming (J. D. Res. **31**: 166, 1951) in the studies of transplanted tooth germs in animals. In addition, Kalnins found that cementicles originate from cementoblasts which have been dislocated from the primary cementum into the periodontium. Kalnins also recognizes the formation of atypical, or pseudo, cementicles which have no connection with the cementoblasts and therefore are not true cementicles.

The bacterial flora of the mouth still represents a difficult area for study. Singer (p. 591) has singled out the oral staphylococci for a consideration. The staphylococci, of course, are commonly present in the oral cavity and show a diversity of biochemical properties with no definite pattern. The effect of urea on the oral staphylococci was investigated and it appears that urea inhibits the toxigenicity of these organisms, or that the alkalinity which results from ureolysis hydrolysis the bacterial metabolites. This basic study of the possible pathogenicity of the staphylococci presents another section in the chapter on oral flora.

The Proceedings of the North-Pacific and the Minnesota Sections are included in this issue. The editor will continue to welcome Sectional Proceedings to the JOURNAL but desires that the abstracts be limited to 200 words each.

Dr. I. R. Sorrie, Secretary-Treasurer of The South African Council for Scientific and Industrial Research, has written to us concerning the desire of the Dental Diseases Sub-Committee of his Council to obtain reprints of articles on dental research published by workers in this field in the United States. The Dental Diseases Sub-Committee of the Council for Scientific and Industrial Research of South Africa is attempting to further programs in research in t h e i r country and has recommended the establishment of a department of dental research in one of the university dental schools of South Africa with the Council contributing financially toward the cost of running the department. Any reprints should be sent directly to Dr. I. R. Sorrie, Secretary-Treasurer, Council for Scientific and Industrial Research, P. O. Box 395, Pretoria, South Africa.

Some of the illustrations used in the article by Doctors Nelsen and Kumpula (*J. D. Res.* **31**: 158, 1952) were reproduced in a trade publication. A check in compensation for this was sent to Dr. Nelsen, who has forwarded it to the JOURNAL OF DENTAL RESEARCH. It is our intention to use this check to support the publication of extra illustrations in the JOURNAL. This generous gesture is greatly appreciated.

H.B.G.R.

INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

Proceedings of the Thirtieth Annual Meeting

Broadmoor Hotel, Colorado Springs, Colo.

March 21, 22, and 23, 1952

Compiled by Hamilton B. G. Robinson, Editor, *College of Dentistry, Ohio State University, Columbus, Ohio*

I. Presidential Address, *Maynard K. Hine (Indiana University)* - - - - - - - 455
II. Index of participants and sequence numbers for abstracts - - - - - - - - 458
III. First Session, Afternoon, March, 21; Abstracts 1-10 - - - - - - - - - - 459
IV. Second Session, Evening, March 21; Abstracts 11-21 - - - - - - - - - - 463
V. Third Session, Morning, March 22; Abstracts 22-33 - - - - - - - - - - 467
VI. Fourth Session, Afternoon, March 22; Abstracts 34-45 - - - - - - - - - 472
VII. Fifth Session, Evening, March 22; Abstracts 46-59 - - - - - - - - - - 477
VIII. Sixth Session, Morning, March 23; Abstracts 60-71 - - - - - - - - - - 482
IX. Seventh Session, Afternoon, March 23; Abstracts 72-95 - - - - - - - - - 487
X. Dental Materials Group; Abstracts 96-104 - - - - - - - - - - - - - - 496
XI. Papers read by title; Abstracts 105-146 - - - - - - - - - - - - - - - 499
XII. Executive Proceedings and register of new members and officers - - - - - 516

I. President's Address: Maynard K. Hine

Indiana University, Indianapolis, Ind.

It has been customary in the past several years for the incoming president of the International Association for Dental Research to be given an opportunity to make an inaugural address. This custom is doubtless based upon the assumption that any man so honored by this Association must have at least one subject upon which he can deliver a somewhat erudite and scholarly discourse. Consequently, immediately after the meeting last year I labeled a manila folder "President's Address" and for several months now have been dropping notes into this file. It is indeed fortunate that the president-elect had but one year to serve, for in another few months I would have needed a filing cabinet to store my notes and a full half-day to deliver my speech.

I want to thank members of the Association for their generous action in granting me the honor of being elected President. I am quick to admit the highest regard for this office, and at the same time, I have a genuine feeling of inadequacy for the position.

Twenty-one years ago, the late Dr. Rickert of Michigan in his presidential address compared the International Association for Dental Research to a young, hopeful "baby." The past years have seen this infant grow and develop into a stable, well-organized, well-respected, and virile adult. Its membership has reached a new high and this year's program includes approximately 100 papers, which is remarkable when one remembers that the meeting place is somewhat distant from the center of the dental school population. In the last score of years the Association has developed qualitatively as well as grown quantitatively. The complexity of some of the reports given at this meeting are actually disconcerting to one who has been merely signing requisitions and salary vouchers

The meetings of the International Association for Dental Research were coordinated with the meetings of the American Association of Dental Schools and Omicron Kappa Upsilon.

The Editor wishes to express his appreciation to Dr. Edward H. Hatton, General Secretary, for cooperation in preparation of these Proceedings.

lately! The philosopher George Santayana once wrote: "Those who speak most of progress measure it by quantity and not by quality; how many people read and write or how many people there are or what is the annual value of their trade; whereas true progress would rather lie in reading or writing fewer and better things . . ." The International Association for Dental Research has grown steadily in size and developed in scope and, if you please, in depth since its organization thirty-two years ago.

In spite of the fact that the program for this year was crowded with research reports, the Program Committee still felt compelled to save time for the President's Address. However, when the American Association of Dental Schools' Program Committee found it necessary to schedule a meeting for Sunday night, the Program Committee of our Association decided to cancel the I.A.D.R. Sunday evening session. This was a wise decision because it will allow everyone in both organizations to listen to discussions of the need of the Armed Forces for manpower rather than a president's address. Consequently, my remarks have been condensed.

I decided last Thanksgiving to discuss with you the science of hermaneutics. I discovered that word in a popular book on archeology and found it to mean "the science of interpretation." The term was coined in 1737 and was applied at that time to the interpretation of the Bible, but gradually usage of the term has broadened its meaning to include interpretation of any subject and sometimes is defined as "the art of not being fooled," certainly an important art for research workers to master. Dental research could be benefited in many ways if some research activities were "interpreted" for the general public by carefully, sympathetically prepared news releases. The American Association for the Advancement of Science has done very well in this regard. Perhaps such a plan can be modified for the I.A.D.R.

In this Thanksgiving version of my president's speech I also discussed the importance of well-controlled laboratory studies but pointed out that carefully interpreted clinical observations should also be accepted as scientific and that we should not be fooled into believing that the only acceptable facts are those that can be verified by the microscope, by chemical analysis, or by a biologic assay. Some problems, such as the value of toothbrushing in the treatment of periodontal disease, or the importance of "foci of infection," cannot be proved easily by direct laboratory studies, but it is unscientfic to throw out carefully-recorded clinical observations by reputable clinicians. Kettering once said that "problems are solved in some fellow's head. They are not solved in a laboratory at all." Of course, laboratory studies are basically important, but we must not scorn the idea from the heads of capable clinicians.

By Christmas time I had decided not to use this speech but to discuss Dental Research—A Catalyst for Dental Education. Any teacher who is involved in an active research program cannot help but be more alert, better informed, and his teaching will be more logical and effective. I am convinced that in one of the better dental schools in the mid-west the level of teaching has improved in part because of the number of sound research projects under way there. This Christmas version of my speech was quite philosophical, and underlined the challenge of dental research to the individual, and the personal satisfactions enjoyed by the individual who energetically pursues research. The value of the results of research to dentistry and the relation of research to the individual's paycheck were to be discussed, as well as the responsibilities of dental administrators to sponsor research by faculty members through giving them adequate quarters, ample time, and effective assistance. This speech I liked when I was tired or drowsy, but, on one of my more energetic mornings, I tore it up because it sounded too much like the speech of a decadent dean.

The speech I finally outlined was a pretty good one, I thought. I discussed the paucity of research in dentistry, particularly in the field of periodontology. In a cursory examination of the 2,155 papers presented at the first 29 meetings of the I.A.D.R., approximately 7 per cent were on periodontia and 13 per cent on caries. I pointed out in no uncertain terms the need for more and better studies of, for example, the physiology of the saliva and the oral structures in general, and I hinted that perhaps one reason this was a neglected field was because of its complexity. I called attention to the fact that one of the most common causes for gingivitis and periodontitis is local irritation due to calculus deposition and yet dental research has produced little evidence regarding the causes of calculus deposition and practically nothing about the methods of prevention of its deposition. Back in 1915, Dr. G. V. Black reported several clinical studies using his ingenious calculus trap and concluded that calculus deposition was, at least in some cases, associated with an excessive diet. To the best of my knowledge this particular theory has never been checked, although it represents one of the best leads we have had for a study of calculus deposition. We should be able to learn how to control the deposition of calcium salts from saliva.

As I said, I thought this speech was pretty good, but in the time I have with you today I cannot give it. Instead I shall comment briefly upon some details of organization of the I.A.D.R.

This Association has as its major objective the sponsoring of dental research, and it sponsors two highly successful projects designed to help meet this objective. These projects, the annual meeting and the Journal of Dental Research, are successful because the I.A.D.R has been fortunate to have capable individuals who are willing to spend the necessary hours and effort to insure their success. We certainly owe our Secretary, Dr. E. H. Hatton and his Assistant, Dr. Dan Burrill, our sincere thanks for their contributions, for they are largely responsible for this meeting. We should also give Editor Hamilton B. G. Robinson credit for carrying forward our second major project, The Journal of Dental Research, which long ago became established as an invaluable force in dental research and is one of the most influential dental journals in the world. The increased size of the Journal has made publication now about as rapid as is consistent with good editing.

The responsibilities for both the Annual Meeting and the Journal are carried by only a few members, leaving many others with talents that should be used. I believe we should undertake some other projects which will help the I.A.D.R. fulfill its objective of promoting "broadly the advancement of research in all branches of dental science." I wish to use my newly granted authority to survey plans for a public relations program, because I am convinced that public interest, and perhaps public funds for research, could eventually be increased by a sympathetic press. Please be assured that this committee will be carefully selected, since we all realize that detrimental publicity is often easier to get than the favorable variety and of course much worse than none at all.

Many of you probably know that the Council on Dental Research of the American Dental Association is preparing to expand its activities greatly in the near future since a full-time executive secretary, who will probably be a member of this Association, is to be chosen soon. As the I.A.D.R. is the most influential organization in this country interested in dental research, it should have an official representative to the Council on Dental Research, to keep the I.A.D.R. members informed of the Council's activities and to help direct the Council's work. I have been assured by the Council on Dental Research that they would welcome a consultant from the I.A.D.R., so our Council voted to

recommend that Dr. Leonard Fosdick be appointed by the Board of Trustees of the American Dental Association as consultant to the Council on Dental Research.

Since it does not seem feasible to hold I.A.D.R. meetings oftener than once a year, it is hoped that there will be more section meetings held during the year. Sections are urged to send reports of their meetings to the Editor of the Journal of Dental Research. Dr. Robinson assures me that they will be welcomed.

In conclusion, may I compliment Dr. Fosdick for his successful year as President, and thank him for his efforts. Thanks also to all who have made contributions to this meeting. Suggestions for improvement of the I.A.D.R's program for next year are solicited; we shall welcome your ideas.

II. Index of Participants, and Sequence Numerals for Corresponding Abstracts

Abel, L. F., 80; Addelston, H. K., 35, 133; Agnew, R. G., 105; Albright, J. T., 64, 65; Andrews, J. C., 84; Armstrong, W. D., 67, 87, 91; Arnold, F. A., Jr., 9; Avant, F. B., 106; Averill, C. J., 106; Avery, J. K., 107; Baker, R., 63; Baume, L. J., 3; Becks, H., 12, 75; Benzer, P., 22; Berkut, M. K., 84; Bernick, S., 63; Bernier, J. L., 142, 146; Beube, F. E., 132; Bhaskar, S. N., 108; Bibby, B. G., 37; Bittner, J. J., 91; Blayney, J. R., 89; Bodecker, C. F., 109, 135; Bogoroch, R., 62; Boozayaangool, R., 10; Boyd, D. A., 2; Boyd, J. D., 55; Boyd, T., 85; Boyle, P. E., 18, 127; Brayshaw, H. A., 45; Broschak, F., 58; Brudevold, F., 38; Burnett, G. W., 16, 26, 60; Burrill, D. Y., 5; Burstone, M. S., 74; Bush, S. H., 100; Cain, E. A., 144; Campbell, R. L., 103; Cannon, J. C., 55; Carter, W. J., 57; Cassidy, P. E., 29; Chernausek, D. S., 110; Chung, P., 103; Claus, E. C., 44; Clausen, H. J., 111; Cohen, A., 95; Colby, J. E., 139; Coolidge, T. B., 47; Costich, E. R., 40; Cox, G. J., 20; Dam, H., 119; DeMarco, C. T., Jr., 116; Detwiler, S. R., 112; Dickson, G., 34; Di Salvo, N. A., 53; Eastman, R. F., 71; Eickenberg, C. F., 31; Ellinger, F., 66; Elliott, H. C., Jr., 56; Engel, M. B., 73; English, J. A., 66; Ennever, J., 113; Epstein, S., 82; Evans, H. M., 75; Ferguson, G. W., 98; Fiasconaro, J., 114, 115, 144; Finn, S. B., 116; Fischer, T. E., 123; Fitzgerald, R. J., 41; Forscher, B. K., 43; Forziati, A. F., 34; Fosdick, L. S., 43, 57, 94, 138; Foss, C. L., 7; Frandsen, A. M., 12, 75; Freeman, F. H., 104; Frisbie, H. E., 19, 117; Fullmer, H., 71; Gardner, D. E., 48; Glasson, G. F., 101; Goldberg, H. J. V., 37; Goldberg, M. A., 118; Granados, H., 119; Greep, R. O., 11; Grupe, H. E., 7; Gurney, G. F., 92; Hadjimarkos, D. M., 120; Hahn, W. E., 106; Harris, M. R., 41; Hawes, R. R., 38; Hein, J. W., 40, 48; Helman, E. Z., 121; Henry, J. L., 59; Hess, W. C., 122; Hester, W. R., 123; Hill, I. N., 89; Hill, T. J., 6; Hoblit, N. E., 97; Horowitz, S. L., 124; Hurst, V., 125; Hutton, W. E., 126; Irving, J. T., 18; Iseri, O. A., 11; Jarabak, J. R., 61, 128, 129, 130, 131, 145; Johansen, E., 81; Johnson, E. L., 15; Johnson, V., 39; Johnston, D. D., 32; Kaiser, W. F., 105; Kamins, M., 129; Karshan, M., 132; Kauffman, S. L., 25; Keyes, P. H., 49; Keyser, J., 79; Klapper, C. E., 52; Klein, E., 35, 133; Koser, S. A., 25; Kraus, F. W., 39; Kreshover, S. J., 72; Kridakara, O., 10; Kutler, B., 88; Kutuzov, H., 1; Kvam, T., 134; Laffre, R. O., 56; Lathrop, D. D., 79; Lee, C., 122; Lefkowitz, W., 53, 135; Leighton, R. E., 55; Levy, B. M., 112; Lindquist, J. T., 96; Lisanti, V. F., 23, 136; Lobene, R. R., 16; Macdonald, J. B., 29; Mahler, I. R., 23; Malone, A. J., 137; Manly, R. S., 77, 78, 79, 80; Mardfin, D., 135; Massler, M., 95, 137; McBride, W. D., 69; McCann, H. G., 93; McClure, F. J., 9, 90; McDonald, R. E., 27; Medak, H., 60; Meyer, J., 59; Mitchell, D. F., 51; Moore, B. M., 57, 138; Mosley, V. M., 64; Munson, P. L., 11; Myers, H. I., 17, 113; Nathans, J., 20; Nelson, M., 75; Nemes, J. L., 24; Neumann, H. H., 53; Newman, M., 6; Noda, L. N., 126; Noyes, H. J., 86; Nuckolls, J., 125; Olson, B. J., 9; Orban, B., 7, 8; Orland,

F. J., 58; Parker, L. W., 11; Perkins, G. T., 139; Pfaffman, C., 79; Philips, R. W., 96; Piez, K. A., 94; Piggott, W. R., 9; Pigman, W., 56; Pincus, P., 140; Pindborg, J. J., 13; Posner, A. S., 36; Ireland, R. L., 88; Irving, J. T., 127; Rae, J. J., 28; Rapp, G. W., 46, 92; Ray, H., 68; Remmert, L. F., 120; Rickles, N. H., 21; Ritchey, B., 8; Robinson, L., 56; Robinson, R. E., 68; Rogosa, M., 30; Rosebury, T., 29; Rushton, M. A., 141; Ryge, G., 99; Saeger, A., 113; Sarnat, B. G., 143; Sausen, R. E., 67; Savara, B. S., 54, 86; Schaffer, A. B., 22; Schlack, C. A., 66; Schoonover, I. C., 34, 98; Schour, I., 59, 95, 108; Schuessler, C. F., 123; Scofield, H. H., 142; Scott, D. B., 64, 65; Scrivener, C. A., 33; Selman, A. J., 143; Shafer, W. G., 70; Shapiro, H. H., 124; Shaw, J. H., 62; Sherman, H., 114, 115, 144; Sherman, R., 68; Shiere, F. R., 77, 136; Sicher, H., 1; Simon, W. J., 67; Sims, J., 6; Singer, L., 87; Skinner, E. W., 97, 103; Smith, D. L., 98; Smith, F. A., 48; Smyd, E. S., 102; Snyder, M. L., 32; Sognnaes, R. F., 62, 65; Spangenberg, H. D., 83; Stephan, R. M., 41; Stephenson, S. R., 36; Storvick, C. A., 120; Strålfors, A., 42; Stuteville, O., 130, 145; Suher, T., 54, 86; Sutton, R. M., 29; Sweeney, E., 62; Sweeney, W. T., 101; Tenebaum, B., 132; Tiecke, R. W., 146; Trautz, O. R., 35, 133; Van Huysen, G., 2, 85; Vehe, K., 131; Vinton, P., 78; Volker, J. F., 10, 50; Weinmann, J. P., 59, 108; West, E. E., 14; Wheateroft, M. G., 24; Williams, N. B., 31; Wirth, L. J., 20; Wiseman, R. F., 30; Wolf, W., 89; Wolfe, W. R., Jr., 5; Wyckoff, R. W. G., 64, 65; Wylie, W. L., 15; Yuktananda, 1, 10; Yurkstas, A., 76; Zander, H. A., 4; Zipkin, I., 90.

III. First Session: Afternoon, March 21; Abstracts 1-10

1. Anatomy and Function of the Palate of the Albino Rat. *Helen Kutuzov and Harry Sicher, Department of Histology, University of Illinois College of Dentistry, and Department of Anatomy and Histology, Loyola University School of Dentistry, Chicago, Ill.* The palates of 25 normal albino rats were examined macroscopically and microscopically with special reference to the functional correlation of their structure to that of the tongue. Four distinct regions, each characterized by a different type of dermal and epithelial structures, were identified: (1) the roof of the oral atrium which carries a longitudinal ridge ending posteriorly at the incisal papilla; (2) the antemolar region, anterior to the molars and characterized by three simple transverse high rugae; (3) the intermolar area, located between and slightly posteriorly to the molars, is occupied by five low ridges in the shape of a letter M straightening out posteriorly to a more simple curve; they carry many sharp secondary ridges; and (4) the postrugal field reaching to the boundary between the hard and soft palate, mainly characterized by innumerable fine papillae. The roof of the oral atrium together with the folding in of the upper and lower lips, and the median sulcus of the tongue form a funnel shaped canal which serves to prevent the entry of not sufficiently comminuted food into the oral cavity proper. The epithelium of the straight rugae contains Merkel's touch cells and, with the underlying portion of the tongue beset with simple conical and fungiform papillae, comprises the discriminatory region of the oral cavity. The sharp secondary ridges of the intermolar area together with the giant papillae of the tongue have an auxiliary masticatory function. The fine papillae of the postrugal field and the filiform papillae of the tongue perform the final screening of the food particles.

2. Tooth Pulp Response to Operative Procedures. *Drexell A. Boyd and Grant Van Huysen, Indiana University School of Dentistry, Indianapolis, Ind.* Eleven cavities in the teeth of dogs were prepared with a steel bur and 10 with the airbrasive technique. Some of the cavities were deep; others shallow. Six

of the cavities were left unfilled; the others were filled with either $Ca(OH)_2$ and cement, cement alone, amalgam, or self-curing resins. One week after the treatment the teeth were removed and semiserial histologic sections prepared. The pulps of the teeth showed histologic evidence of injury only if the cavities were unfilled. This is true regardless of whether or not the cavity was deep or shallow, prepared with a bur or airbrasive technique.

3. Observations Concerning the Histogenesis of the Epithelial Attachment. *L. J. Baume, D.M.D., M.S., Division of Dental Medicine, College of Dentistry, University of California, San Francisco, Calif.* The development of the epithelial attachment and the formation of a gingival sulcus was studied histologically in the first permanent molars and the incisors of 20 Rhesus Macaques at consecutive preeruptive and posteruptive stages. Following the formation of the enamel there occurred a reduction of the elements of the stratum intermedium, stellate reticulum and outer enamel epithelium into a syncytium of uniform prickle cells. Their cytoplasm stained only lightly with fuchsin (Masson's stain) while that of the oral epithelium always stained much darker, allowing a clear differentiation, especially under the phase contrast microscope. The prickle cells were connected with the ameloblasts by intercellular bridges containing prominent tonofibrils. Near the surface of the enamel these tonofibrils fused together to form a continuous membrane, which eventually calcified. In the pre-eruptive stage the following processes were observed: (a) Catabolic changes in the enamel organ, (b) Desmolysis of the connective tissue between the oral and enamel epithelium, (c) Fusion of the oral and enamel epithelium. There was a variation in the timing of these events apparently related to the stresses exerted by the horizontal and vertical movements of the erupting tooth: (1) At the sites of eruptive pressure the prickle cells of the enamel epithelium proliferated interstitially and replaced the ameloblasts prior to clinical tooth eruption. At the same time oral epithelium proliferated toward the enamel epithelium, fused with it and grew along the basal layer of the latter. After the tip of the crown had emerged the enamel epithelium began to disintegrate at the points of fusion of the two epithelia. This established a sulcus between remnants of the reduced enamel epithelium and oral epithelium. (2) In the absence of pressure at the opposite side the prickle cells proliferated at their basal side in the presence of intact ameloblasts. Oral epithelium did not fuse immediately with the enamel epithelium, but proliferated first some distance behind it and followed later the pattern described under (1). Studies with the phase contrast microscope show that even though the two epithelia fuse the enamel epithelium does not change into stratified squamous epithelium by metaplasia. There was evidence that a normal physiologic desmolysis was the precursor of the proliferation of the oral epithelium and thus controls the downward growth of cells of the epithelial attachment.

4. Calculus Attachment to Cementum. *H. A. Zander, School of Dentistry, University of Minnesota, Minneapolis, Minn.* The removal of calculus from root surfaces is sometimes easy and sometimes very difficult, with many variations in between these two extremes. While this has been thought to be due to variations in the hardness of calculus, it is possible that the organic connection between tooth and calculus may have something to do with it. Therefore, decalcified and ground sections were prepared, stained, and examined from autopsy specimens and freshly extracted teeth. Four variations in modes of attachment have been demonstrated so far: (1) the organic matrix of calculus is attached to the secondary cuticle; (2) the cuticle is absent and the calculus matrix is attached to minute irregularities in the cementum surface; (3) organisms have penetrated into the cementum and are continuous with the organisms

in the calculus matrix; (4) calculus is locked mechanically in undercut areas of cement resorption. It is suggested that these differences in attachment may account for differences in the difficulty or ease with which calculus is removed and may point toward reasons why calculus reoccurs faster on some teeth in some mouths than in others.

5. Effect of a Penicillin Dentifrice on Gingivitis. *W. R. Wolfe, Jr., and Dan Y. Burrill, School of Dentistry, University of Louisville, Louisville, Ky.* The gingival health of 201 subjects was evaluated by charting all areas of inflammation about the teeth, as shown by hyperemia, exudation, edema or cyanosis. Half of the group, selected at random, was issued a powder dentifrice containing 500 units of penicillin per gram, and the other half was issued the same dentifrice without the penicillin. After 6 months the 100 control subjects showed an average increase of 4.20 areas of inflammation (from 12.00 to 16.20) while the 101 subjects using the penicillin dentifrice showed an average increase of 2.02 areas (from 14.07 to 16.99). Fifty-five of the subjects were followed for one year. Of these, the 29 controls showed an average increase of 3.12 areas (from 10.00 to 13.12) and the 26 using the penicillin dentifrice showed practically no change (from 12.15 to 12.10). Forty-six of the subjects were given dental prophylaxis one week before the initial examinations. The average increase of inflamed areas for this group in 18 months was as follows: 7.43 areas (from 9.20 to 16.63) for the 22 controls and 9.96 areas (from 7.54 to 17.50) for the 24 subjects using the penicillin dentifrice. The increases in numbers of inflamed areas are not explained. It is concluded that the effects of the penicillin dentifrice used were not clinically significant.

6. The Effect of Penicillin Dentifrice on the Control of Dental Caries and the Production of Resistant Organisms. *Thomas J. Hill, John Sims, and Millicent Newman, School of Dentistry, Western Reserve University, Cleveland, Ohio.* An experiment using penicillin dentifrice was conducted in the schools of South Euclid, Ohio. Four hundred and ninety-two children from ages 7 to 12, inclusive, used a penicillin-containing paste for one year. The program was regimented and the teeth were brushed once a day in the school and pupils urged to have additional brushings at home. Results at the end of the year showed that there was a very slight reduction in dental caries. This reduction was in the following range: decayed-missing-filled teeth, 9.74 per cent; decayed-missing-filled surfaces, 15.93 per cent; total lesions (cavities and fillings), 14.52 per cent. Only the reduction in decayed-missing-filled surfaces was statistically significant. There was no important change in lactobacillus counts during the year. There was an increase in the number of penicillin resistant forms of *Streptococcus viridans.*

7. Gingivosis. *Calvin L. Foss (San Diego, Calif.), Harold E. Grupe (Amarillo, Texas), Balint Orban (Colorado Springs, Colo.), Colorado Dental Foundation, Colorado Springs, Colo.* Gingivosis is characterized by extensive degenerative changes in the connective tissue as well as in the epithelium of the gingiva. The disease is not localized to the free margin of the gingiva but extends to, or is localized to, the attached part of the gingiva. Ten cases have been studied by histopathologic method. The changes in the connective tissue are disintegration (depolymerization) of the ground or cementing substance with consequent breakdown of collagenous fiber bundles. These changes are accompanied by an extensive inflammatory reaction which is secondary. The epithelial changes are degeneration of the intercellular substance as well as cells. Hyalin degeneration is frequently observed. Atrophy of the epithelium occurs, and the epithelium is often separated from the underlying connective tissue. This separa-

tion of the epithelium was the reason that certain types of the disease were previously termed desquamative gingivitis. However, there is no true desquamation present and the gingivitis is secondary. The primary feature of this disease is degeneration.

8. Cysts in the Gingiva. *Beryl Ritchey and Balint Orban, Colorado Dental Foundation, Colorado Springs, Colo.* Cysts in the gingiva may not occur as infrequently as could be assumed from the literature. This report includes five cases. The etiology is thought to be: (1) invagination of gingival epithelium, (2) traumatic displacement of epithelial cells, (3) epithelial remanents of the dental organ, or (4) remanents of ectopic gland structures. Gingival cysts may never produce clinical signs or symptoms; by their continuous enlargement they may produce mechanical interference; and, they may become infected and develop into an acute abscess. Clinical diagnosis should exclude simple ulcers and periapical lesions or periodontal abscesses. Treatment of choice is complete excision.

9. Studies of Fluorine in Experimental Tuberculosis. *B. J. Olson, F. J. McClure, W. R. Piggott, and F. A. Arnold, Jr., The National Microbiological Institute, and The National Institute of Dental Research, National Institutes of Health, Public Health Service, Federal Security Agency, Bethesda, Md.* The epidemiologic studies of fluoride-bearing drinking waters have demonstrated that at certain concentrations of fluorine there is marked inhibition of dental caries. This affords some explanation on the known difference in geographic distribution of dental caries in a population. The possible effect of this water borne trace element on other diseases with known geographical variations such as tuberculosis, needed investigation.

Studies of orally administrated fluorine to guinea pigs infected with experimental tuberculosis yielded inconclusive results, necessitating studies in other animals. Dosages of 2.0 mg. and 0.1 mg. of fluorine/kg. of body weight given monkeys orally as sodium fluoride, did not adversely affect their survival time as compared with monkeys given 2.0 mg. of chlorine/kg. orally as sodium chloride. All the monkeys were infected with 0.0001 mg. of a virulent human strain of tuberculosis. In this preliminary experiment the monkeys receiving 0.1 mg. fluorine/kg. daily, as sodium fluoride survived longer than the monkeys in the other 2 groups. This interesting preliminary experimental result in monkeys necessitates further investigation and confirmation.

10. Dental Survey of Selected Thai Children. *Od Kridakara, Rith Boozayaangool, and Isra Yuktananda, Dental School, University of Medical Sciences, Bangkok, Thailand, and J. F. Volker, University of Alabama School of Dentistry, Birmingham, Ala.* A clinical survey of the teeth and supporting structures of over 3,000 Thai children has been made. Approximately half this number were from the captial city of Bangkok, located in the south central portion of the country. The remainder were from the cities of Chiengmai and Lampang in the northern hill country. It should be noted that Bangkok has considerable personal and commercial contact with western civilization while Chiengmai and Lampang are relatively isolated. The deciduous teeth of Bangkok children showed extensive dental caries. At the age of 4, the average deciduous DMF is approximately 10. Caries of the permanent teeth is relatively moderate. The average permanent DMF at age 18 is 5.5. In contrast, caries of the deciduous teeth of Chiengmai children is of limited severity. At the age of 6, the average deciduous DMF is 3. The permanent teeth of Chiengmai children show an even greater degree of caries freedom. At the age of 18, the average DMF is 1.5. The caries attack rate in Lampang parallels that of

Chiengmai. The Bangkok area showed the greatest number of children needing orthodontic treatment and residents of Chiengmai the least. The reverse relationship was found regarding the need for gingival therapy. Figures from government sources indicate that the per capita consumption of refined carbohydrates in increasing rapidly. This is particularly true in the Bangkok area and very probably is responsible for the increased caries attack rate.

IV. Second Session: Evening, March 21; Abstracts 11-21

11. The Acute Effect of Parathyroidectomy on the Serum Calcium and Inorganic Phosphorus of Rats Maintained on a Low Calcium Diet. *Paul L. Munson, Oscar A. Iseri, Lorelei Waite Parker, and Roy O. Greep, Biological Research Laboratories, Harvard School of Dental Medicine, Boston, Mass.* Male rats of the Sprague-Dawley strain were kept on a diet essentially free of calcium for 1 to 3 weeks, beginning at 28 days of age. The rats were then parathyroidectomized by a new rapid technique which permits complete extirpation with minimum trauma and minimum loss of blood. Under ether anesthesia the parathyroids are exposed and, under a dissecting microscope, are destroyed by electrocautery. Within 4 to 6 hours after the operation the serum calcium dropped from the preoperative level of 9 mg. per cent to an average of 5 mg. per cent. During the same time interval no significant change in serum inorganic phosphorus could be detected. Subcutaneous administration of 100 U.S.P. units of commercial parathyroid extract immediately following parathyroidectomy partially or completely prevented both the decline in serum calcium and the accompanying tetany. These results suggest that in the rat the primary effect of parathyroid hormone is on calcium metabolism and not on phosphorus metabolism as has been widely believed. The effect of parathyroid hormone on the acute decline in serum calcium following parathyroidectomy in the calcium-depleted rat is being investigated as a possible convenient biologic assay method.

12. The Influence of Pantothenic Acid and Vitamin A Deficiencies on the Healing Process Following Molar Extraction in the Rat. *Asger M. Frandsen, D.D.S., Hermann Becks, M.D., D.D.S., Division of Dental Medicine, College of Dentistry, University of California, San Francisco, Calif.* Three groups of animals were studied: (I) 33 male rats raised on a stock diet. (II) 35 male rats raised from the age of 30 days on a purified diet deficient in pantothenic acid. (III) 28 male rats raised from the age of 21 days on a purified diet deficient in vitamin A. The experimental animals of the last two groups had pair-fed controls. All animals had the extraction performed at the age of 60 days, and they were then sacrificed after various postoperative intervals ranging from 3 hours to 40 days. The socket area was sectioned buccolingually in serial sections and stained with hematoxylin and eosin. In the first group initial formation of new bone took place 5 days after the extraction. Epithelial closure was established between 8 and 10 days of healing. Twenty-five days after the extraction the socket was completely filled with new bone. In the second group the sockets of both experimental and pair-fed control animals were about half filled with new bone 5 days after the extraction. At this time, too, epithelial healing had taken place in both categories of rats. Twenty-five to 30 days after the extraction the sockets of both deficient and control animals were filled with bone and reorganization of the trabecular arrangement had begun. The rats of this group exhibited the typical gross symptoms of pantothenic acid deficiency. In the third group the sockets of both experimental and pair-fed control animals were about half filled with new bone 5 days after the extraction, and at the same time epithelial healing was established. Twenty-

five to 30 days after the extraction the sockets of both deficient and control animals were filled with bone and reorganization had begun. The rats in this group exhibited gross symptoms of vitamin A deficiency and, on histologic examination, the characteristically changed incisors. As far as the rate of bone healing and epithelial closure is concerned the purified diets proved superior to the stock diet. The degree of pantothenic acid and vitamin A depletion produced in this experiment did not appear to influence the healing process following extraction.

13. The Effect of Antabuse Upon Dental Changes in Vitamin E-Deficient Rats. *J. J. Pindborg, D.D.S., Dr. Odont., Departments of Dental Histology, University of Illinois, and the Royal Dental College, Denmark and Medicinalco, Ltd., Copenhagen.* Vitamin E-deficiency prevents the formation of pigment in the rat incisor. Aaes-Jorgensen and associates (*Acta. pharmacol. et toxicol.* 7: 171, 1951) showed that addition of antabuse to a vitamin E-deficient diet protects against the loss of pigmentation. This finding was confirmed in a new series of experiments and was extended by a histologic examination. Twenty 1-month-old rats were placed on a vitamin E-deficient diet for 4 months. During this time the incisors showed a partial lack in pigmentation. Ten rats were continued on the original diet for an additional 3 months (Group I). They showed a further increase in the lack of pigmentation. The remaining 10 rats received, for a similar period of 3 months, 0.025 per cent antabuse mixed into the experimental diet (Group II). They had normal pigmentation of all their incisors. After the 3 months the histologic examination revealed in Group I the typical changes in the enamel organ of the vitamin E-deficient rats; premature atrophy of the papillary layer, edema, and formation of cystlike cavities. In contrast all the animals in Group II exhibited normal conditions in the enamel organ. From this study it appears that antabuse can, as far as the rat incisor is concerned, fully replace vitamin E.

14. Facial Pattern in Malocclusion. *Eugene E. West, B.S., D.D.S., University of California, College of Dentistry, San Francisco, Calif.* Downs has developed a method for the analysis of facial pattern. His standards are based on studies made on a group of individuals possessing excellent occlusion. This study was made to determine how Class I and Class II, Division I malocclusions (Angle) would differ from the group with which he worked. Accordingly, headfilms of 79 Class I malocclusions and 123 Class II, Division I were selected from the files of the orthodontic department. These were analyzed using the method of Downs. The results show that there was the expected significant differences between the excellent occlusions and the malocclusions. The difference, however, between the Class I and the Class II, Division 1 malocclusions was not nearly as marked as might have been expected. The skeletal patterns of the two, with the exception of the angle of convexity and the A-B facial plane angle, were not as marked as we would expect if we were simply using the Angle classification. In the denture pattern (tooth relationships) the differences were more as one would expect to find them with the exception of the angles found by projections from the long axis of the lower incisor to the occlusal plane and lower incisor to the mandibular plane. This would suggest that when we discuss malocclusions by the Angle classification, particularly Class II, Division 1, that many times we are attributing to an individual a facial pattern that is much poorer than he actually has. The facial pattern is much better described by using terminology from the Downs analysis.

15. Rapid Evaluation of Facial Dysplasia in the Vertical Plane. *Wendell L. Wylie, D.D.S., M.S., and Ernest L. Johnson, D.D.S., University of*

California, College of Dentistry, San Francisco, Calif. Orthodontists often speak of "good" and "poor" facial patterns, usually without defining the distinction in quantitative terms, although the Frankfort mandibular plane angle and other angles are coming into increasing use in this connection. Because angles serve poorly to localize and differentiate, this study is directed at showing specifically how certain anatomical areas vary when esthetic distinctions are drawn. Lateral headfilms taken with the teeth in occlusion provided the basis for this study; 171 of these taken prior to orthodontic treatment in an age group of 11 to 13 years were segregated into 57 "good," 61 "fair," and 53 "poor," using subjective appraisal only. On each film measurements of facial height at the profile, length of the mandibular body and the mandibular ramus were made, the gonial angle was measured and the vertical placement of the glenoid fossa of the temporal bone was determined; differences between means were evaluated for statistical significance. The data show that subjective evaluation tends toward "poor" when: lower face height becomes large, when ramus height becomes short, when the angle of the mandible becomes large, and when placement of the glenoid fossa of the temporal bone is relatively high. The end product of this study is a set of transparencies for the assessment of vertical dysplasia from lateral films, so that each individual may be placed in relation to the rest of the population with respect to a given variable) without tracings and without actual measurements being required.

16. The Chemical Composition of Syrian Hamster's Teeth. *Ralph R. Lobene and George W. Burnett, Dept. Dental Research, Dental Division, Army Medical Service Graduate School, Washington 12, D. C.* An investigation has been made of the chemical composition of Syrian hamster's teeth by spectrographic and standard analytical procedures. The teeth of 30 sacrificed animals were separated into collective groups of incisors and molars and extracted with ether and ethyl alcohol. Each group was ground to 100 mesh particle size. Enamel and dentin were then separated by floatation methods. Spectrographic analysis of isolated incisor enamel showed less than 0.01 per cent silver, aluminum, manganese, lead, and strontium; from 0.01 to 1 per cent copper, iron, magnesium, sodium, silicon, and zinc; and more than 1 per cent calcium and phosphorus. Spectrographic analysis of isolated incisor dentin showed less than 0.01 per cent silver, aluminum, boron, barium, manganese, lead, and strontium; from 0.01 to 1 per cent copper, iron, sodium, and silicon; and more than 1 per cent calcium, phosphorus, and magnesium. The organic content of incisor enamel was less than 3 per cent; that of incisor dentin averaged 31.17 per cent. The average calcium and phosphorus content of ashed incisor enamel was 39.49 per cent and 19.46 per cent respectively. The average calcium and phosphorus content of ashed dentin was 35.10 per cent and 19.38 per cent respectively. The calcium-phosphorus ratio of incisor enamel was 2.03; that of incisor dentin was 1.81. Similar studies of the composition of hamster's molar teeth are being made for comparison with the composition of the incisors.

17. The Distribution of Osteoclasts in the Mandibular Joint of the Guinea Pig. *Hugh I. Myers, University of Kansas City, School of Dentistry, Kansas City, Mo.* The material used in this study was taken from male guinea pigs of known ages and weights. The mandibular joints were removed intact, fixed in 4 per cent formalin, decalcified, and imbedded in paraffin. Serial sections at 6 μ were made and every tenth section mounted and stained with hemotoxylin and eosin. It was found that the distribution of the osteoclasts indicated a relationship to the known pattern of mandibular condyle growth. Almost all the

osteoclasts were found in the following regions: (1) In the marrow spaces adjacent to the cartilage layer—indicating a role in its destruction; (2) In the marrow spaces adjacent to the bone spicules in the upper ramus and neck—indicative of the change from the cartilage-containing spicules of the neck to the relatively dense bone of the ramus; (3) In the periosteum of the neck adjacent to the surface bone—indicative of the marked bone destruction necessary to convert the knob-shaped condyle to the smaller neck and flat ramus.

18. Occurrence of Mitotic Figures Among Odontoblasts and Other Cells of the Dental Pulp in the Teeth of Scorbutic Guinea Pigs. *Paul E. Boyle and James T. Irving, School of Dentistry, University of Pennsylvania, Philadelphia, Pa.* Sections of the incisor and molar teeth of animals in various stages of vitamin C deficiency and repair have been examined. The occurrence of mitotic figures among the odontoblasts and adjacent pulp cells was noted in well-developed acute scurvy. They were particularly abundant in areas where normally dentin formation is at a peak, but where in the deficient animals dentin deposition had essentially stopped. Proliferation of cells appears to occur as a compensatory phenomenon where normal differentiation and function of the cells is inhibited. It is concluded that odontoblasts are "reverting post-mitotic" cells. This may explain the formation of new dentin after pulpotomy by wandering odontoblasts or the decendants of mitotic division of odontoblasts.

19. Terminal Dentinal Tubules and Tubules Appearing to Cross the Dentinoenamel Junction. (Enamel Spindles). *Harry E. Frisbie, University of California College of Dentistry, San Francisco, Calif.* Using ground sections, the histologic arrangement of the peripheral dentinal tubules was studied by axial transmitted lighting and by phase contrast microscopy. Arborization into many fine parallel branches which are closely placed and terminate freely at essentially right angles to the dentinoenamel junction correspond to the appearance in young developing teeth as previously reported. Occasional terminal tubules are of much greater diameter than the majority. Numerous shadows which demarcate the dentinoenamel junction at different levels are seen due to the obliqueness of this junction in the relatively thick section obtained by grinding. This is further complicated by the concavities and their peripheral projections and peaks at the dentinal surface. Tubules which at one focus appear to project as spindles into the enamel may at a changed focus be shown to terminate within the dentin limits. Extremely long spindles have been found to be composed of long narrow areas of dentin which may include a number of fine terminal tubules in addition to the principal larger one. The microscopic picture often becomes further confused by a spiral tuft of enamel rods projecting out from this area into the enamel structure. The overall effect may produce varishaped spindle appearing structures.

20. A Method of Study of Molar Form in the Rat. *Gerald J. Cox, Jessica Nathans, and Leo J. Wirth, School of Dentistry, University of Pittsburgh, Pittsburgh, Pa.* Rat molars exhibit variations in (a) subdivision of major cusps, (b) fusion of minor to major cusps, (c) absence of minor cusps, (d) presence of extra cusps, (e) and bifurcation of minor cusps. Some of these variations are related, in frequency, to the diet of the mother during pregnancy and lactation; others may be related to familial, and hereditary factors. A method of describing and of scoring 32 such features, and their bilateral occurrences, has been developed for recording on IBM cards for analysis of the relations to diet, family, and strain, and for exploring interrelationships. In some cases

unusual whole tooth forms must be described since they occur in low frequency. Provision is made for recording the angle of rotation of the third upper molar about the distal-mesial axis of the maxillary teeth.

21. The Estimation of Dental Caries Activity by a New Colorimetric Test. *Norman H. Rickles, D.D.S., M.S., Division of Dental Medicine, College of Dentistry, University of California, San Francisco 22, Calif., and Dental Corps, Percy Jones Army Hospital, Battle Creek, Mich.* It was the purpose of this investigation to evaluate: (1) The accuracy and practicability of a new colorimetric test for the determination of dental caries activity and future dental caries increment, (2) the degree of correlation of the L. a. index and total titratable acid (of an incubated saliva-sucrose mixture) with the dental caries increment, and (3) the relationships which may exist among the three tests. Clinical and bite-wing roentgenographic dental examinations were made of 135 students (mean age, 22); the findings were recorded according to the modified Boedecker dental caries index. Early morning saliva samples were collected for the following determinations: (1) The pH of each of the saliva samples, when mixed and incubated with a sucroindicator solution and compared with standard color tubes, (2) the total titratable acid of each of the above mixtures, and (3) the L. a. index. One year later the dental examinations were repeated under identical conditions, plotted against the initial laboratory tests and subjected to statistical analysis: (1) The lower the pH of the saliva-sucrose mixture, the more dental caries occurred during the ensuing year, (2) the more total titratable acid produced, the greater the dental caries increment, (3) the dental caries increment could be predicted (within definite limits) for the higher the L. a. index, the greater the increase in dental caries, (4) individual dental caries increment could be predicted (within definite limits) for the following year if no dietary changes were made, (5) the new colorimetric pH test and the determination of total titratable acid were equally accurate in their prognostic abilities.

V. Third Session: Morning, March 22; Abstracts 22-33

22. The Use of Hyaluronidase in the Treatment of Traumatic Swellings. *Paul Benzer and Arnold B. Schaffer, Department of Oral Surgery of New York University College of Dentistry and Bellevue Hospital, New York, N. Y.* A series of 50 surgical and nonsurgical traumatic swellings of the head and neck which ordinarily would have indurated and persisted from 1 to 3 weeks was treated with hyaluronidase, an enzyme which has been shown to lyse the ground substance of the connective tissue. As a result of this therapy there is a rapid diffusion of accumulated fluid away from the area of lymphedema and swelling. One hundred and fifty turbidity reducing units of hyaluronidase dissolved in 3 to 10 c.c. of physiological saline was introduced directly into the swellings. The amount of saline used is that quantity that is necessary to raise the hydrostatic pressure in the area. Hematomas, due to injection and trauma, so treated underwent a reduction of 80 per cent in size within 30 to 60 minutes. Reductions in the size of postsurgical swellings were similarly accomplished but with less magnitude. A significant result in all cases was a diminution of the subjective symptoms of tension and pressure. Similarly pain was decreased in the area. Trismus in many cases was partially or totally relieved. Objectively there was a decrease in the size of the swelling and a reduction of induration. Cases so treated healed in a shorter time than would have occurred if they had not received this therapy. The nature of the response of the swellings to hyaluronidase will be analyzed in terms of our existing concepts of inflammation and the mechanism of the action of hyaluronidase.

23. The Significance of the Variation of Hyaluronidase Activity in Human Saliva. *V. F. Lisanti and I. R. Mahler, Tufts College Dental School, Boston, Mass.* The presence of certain oral diseases has been shown to elevate the hyaluronidase activity of human saliva. In order to evaluate the significance of this elevation, titers for this enzyme have been determined on 173 subjects from 3 to 34 years of age. Good correlation was obtained with the number of decayed teeth significant to the 1 per cent level. Periodontal lesions, whether caries were present or not, caused an increase in hyaluronidase activity in 17 cases examined. Sixty subjects having colds or upper respiratory involvements demonstrated 2.5 times the activity of the 95 controls. Thirty of these subjects were tested before and during colds or upper respiratory disturbances. During the active symptomatic phase of these disturbances, the enzyme activity was 3.2 times the level when free of disease. Statistical evaluation of the data indicates the probability that findings are extremely significant below the 1 per cent level.

24. Demonstrations of Hyaluronidase in Saliva by the Streptococcal Decapulation Test. *M. G. Wheatcroft and J. L. Nemes, Naval Medical Research Institute, Bethesda, Md.* Salivas of 50 individuals have been tested for the presence of hyaluronidase. A test has been devised employing encapsulated Group C mucoid beta hemolytic streptococci as the substrate (Murray and Pearce, *Canad. J. Res.* 37: 254, 1949.) One of the components of the capsular material is hyaluronic acid. The enzyme hyaluronidase causes a depolymerization and hydrolysis of the capsular material resulting in the gradual disappearance of the capsular zone. This test is reported to be more sensitive than other tests employed in the past. Turbidity or viscosity of the saliva do not interfere with the results. Of the 50 individuals tested by this method all but one showed the presence of hyaluronidase in the saliva.

25. The Amount of Folic Acid in Saliva. *Stewart A. Koser and Shirley L. Kauffman, Zoller Dental Clinic and Department of Bacteriology and Parasitology, University of Chicago, Chicago, Ill.* By microbiologic assay it was found that the amount of free folic acid (pteroylglutamic acid) plus any substances with folic acid activity averaged 0.024 micrograms per ml. of saliva. This figure was the average of 51 separate determinations on saliva specimens from 20 persons, mostly young adults. Most of the values fell between 0.008 and 0.039 micrograms per ml., with extremes of 0.003 to 0.075 micrograms per ml. In testing the accuracy of the assay method, satisfactorily uniform values could be obtained on different amounts of the same saliva specimen, and folic acid added in known amounts to specimens could be detected within the usual limits of accuracy of a microbiologic method.

26. The Respiration Rate of Human Saliva. *George W. Burnett, Department of Dental Research, Army Medical Service Graduate School, Washington 12, D. C.* The respiration rate of stimulated saliva from 34 humans has been determined by conventional manometric techniques. The relationship of the respiration rate to lactobacilli counts, acid producing potential, amylase activity and fluorides was investigated. The respiration rate of saliva from different individuals varied over a wide range but was relatively constant for each individual from week to week. The respiration rate of whole stimulated saliva was not correlated with lactobacilli counts but, in several cases, a high respiration rate was associated with freedom from caries. The addition of mono-, di-, and polysaccharides to saliva in vitro increased low respiration rates more than it did high respiration rates. The acid producing capacity of various salivas varied

considerably when acting on different carbohydrates. Many salivas produced more acid from polysaccharides (starch) than from mono- and disaccharides. This phenomenon was not directly correlated with the basic respiration rate but apparently depended, in part, upon the amylolytic activity of the individual saliva. Low concentrations (5 parts per million or less) of various simple and complex fluorides did not appreciably lower the respiration rate of saliva in vitro. Higher concentrations of fluorides (200 parts per million) lowered the basic respiration rate of saliva but did not suppress it. Fluorides lowered the respiration rate of saliva to which carbohydrates had been added more than it did saliva to which no carbohydrates had been added.

27. The Effect of Antihistaminic Drugs on Salivary Flow and Viscosity. *Ralph E. McDonald, School of Dentistry, Indiana University, Indianapolis, Ind.* Antihistaminics have been found to have action similar to atropine and a parasympathetic blocking effect, in addition to their detoxifying power against histamine. The effect of minimal effective doses of Benadryl, Pyribenzamine, Clor-Trimeton, and Thephorin was observed on the salivary flow and viscosity of 7 adults. After each individual's normal salivary flow and viscosity had been determined, the reaction to each drug was observed over a 5 day period. Benadryl, Pyribenzamine and Clor-Trimeton depressed the salivary flow in all of the subjects. Clor-Trimeton exhibited the greatest depressant action, a 25 per cent reduction in flow for the group of subjects. Benadryl and Pyribenzamine each brought about an average 20 per cent reduction in flow for the group. Greatest reduction in flow for any one individual on a given day was 54 per cent for Clor-Trimeton and 50 per cent for Pyribenzamine. Benadryl caused the greatest average increase in viscosity, 8.7 per cent, with some subjects showing as much as a 33 per cent increase. A different reaction to minimal effective doses of Thephorin was noted. The salivary flow was increased an average of 14 per cent in the group, with individual increases as high as 36 per cent. Thephorin reduced salivary viscosity in all but one case; the greatest reduction observed was 15 per cent. Since some of the antihistaminics have been shown to depress salivary flow and increase viscosity, it is possible that prolonged use of the drugs may depress salivary action and increase the viscosity to the extent that an increase in dental caries as well as other oral manifestations may be noted.

28. Ammonia Production in Saliva. *James J. Rae, University of Toronto, Toronto, Canada.* In a previous report it was found that the ammonia producing ability of saliva reached a maximum in 24 to 48 hours on incubation at 37°; was independent of whether the saliva was stimulated or unstimulated; was inhibited by sodium fluoride; was irreversibly activated on heating to 50° for 24 hours and was removed by filtering the saliva through a Seitz filter or a fritted-glass funnel (*J. D. Res.* **30**: 385, 1951). The present investigation extended this study to 66 salivas which were divided into 3 groups on the basis of lactobacillus counts. It was found that the activity of the ammonia producing mechanism was unrelated to the lactobacillus count. The urease content of saliva was also found to be unrelated to the lactobacillus count. The relation between urease activity and ammonia production in saliva was not a significant one. In all cases the presence of 2.5 per cent glucose was found to stimulate the ammonia production in the presence of urea.

29. A Study of the Motile Anaerobic Rods of the Oral Cavity. *John B. Macdonald, Ruth M. Sutton, and Pauline E. Cassidy, Faculty of Dentistry, University of Toronto, Toronto, Canada, and Theodor Rosebury, School of*

Dentistry, Washington University, St. Louis Mo. Seventeen strains of motile anaerobic rods have been isolated from the gingivas of mouths showing varying degrees of gingival inflammation. Experiments have dealt with isolation, cultivation and characteristics of these motile anaerobes. Methods investigated for the isolation of these organisms have included serial cultivation of exudate in fluid media, bacteriostasis with the use of 3 different dyes in various concentrations, satellite formation on blood agar and finally alternate cultivation in fluid media and on blood agar from the migrating edge of confluent growth on blood agar or in spirochete cup plates. Of these, only the last method has been successful. Observation of the characteristics of the forms isolated indicate that they include all the morphologic forms normally observed in guinea pig fusospirochetal exudate and that they may be classified into 3 groups. Group I consists of slender actively motile darting vibrating rods which on blood agar produce low convex gray white colonies surrounded by a zone of greening. They produce only scanty growth in highly complex fluid media. Group II consists of slender "fusiform" rods and long filaments. The cells are flexible and show jerky alterations in contour from straight to curved or bent outlines and back again to straight. Lashing movements around a fixed pole are also seen. Movements of translation are very rare. Members of this group produce gray depressions in blood agar. They grow well in a variety of fluid media but show motility rarely except in veal heart ascites broth with added guinea pig kidney extract. Group III consists of thicker curved rods or short spirals showing a sub polar flagellum on the concave surface and exhibiting a rapid tumbling motility. They grow profusely in Difco thioglycollate broth and very poorly or not at all in all other fluid media tried. This group does not grow on the surface of blood agar plates. There are minor but consistent differences in the biochemical activity of the three groups. None are pathogenic subcutaneously in guinea pigs.

30. A Colorimetric Test for the Relative Abundance and Acidogenic Potential of Lactobacilli in Saliva. *Morrison Rogosa and Ralph F. Wiseman, National Institute of Dental Research, Bethesda, Md.* Based on the selective lactobacillus medium (SL Agar), of Rogosa, Mitchell, and Wiseman (*J. D. Res.* **30**: 682, 1951), a technique was developed for the determination of (1) the relative abundance of lactobacilli in saliva, or (2) their acidogenic potential, or (3) both. The foregoing medium was modified by the addition of 0.05 per cent 1(+) arabinose; 0.004 per cent alizarine red S (sodium alizarine sulfonate); and also by the reduction of the agar concentration to 0.2 per cent. The medium was heated minimally to dissolve the agar and was dispensed in 5 ml. quantities into 16 × 150 mm. tubes. Further heating was disadvantageous, since the indicator exhibited undesirable change. The tests were performed repeatedly during 1 year on the salivas from 150 individuals. One loopful of saliva was inoculated into each tube and incubated at 37° C. for 48 hours. The color changes were read after 24 and 48 hours. A change from purple to yellow was considered positive. Based on the rate and extent of color change, a scoring system from 1 to 10 (1 being negative and 10 most positive) was correlated with the lactobacillus counts from SL agar which were scored 0-1000=1; 1000-10,000=2; 10,000-100,000=3; 100,000-1,000,000=4; >1,000,000=5. Correlation of the results between the 2 procedures was +0.9988 with a scatter, or standard error of estimate, of 0.0874.

31. Effects of Sonic Vibration on the Numbers of Microorganisms Cultivable From Human Saliva. *Ned B. Williams and C. F. Eickenberg, Department of Microbiology, School of Dentistry, University of Pennsylvania,*

Philadelphia, Pa. A series of studies were carried out to compare the effects of mechanical shaking and sonic vibration on the numbers of cultivable micro-organisms in saliva. Saliva was collected from 4 human beings, pooled and divided, one portion was treated on a shaker delivering about 85 cycles/second and the second portion was exposed to sonic vibration at a freuency of 9,000 cycles per second, power output of 50 watts. Standard 1.0 ml. aliquots were removed at varying time intervals diluted and inoculated on blood agar in Petri plates. The plates were incubated aerobically at 37° C. for 48 hours, then all colonies appearing on the plates were counted. It was found that the counts from the 2, 6, 15 and 30 minute aliquots from the sample treated by sonic vibration were 5 times higher, and the count from the 10 minute aliquot was 10 times higher than the counts from similar aliquots of the sample treated by mechanical shaking. The counts from all aliquots of the sample treated by mechanical shaking were not markedly increased even after 30 minutes. Different lots of pooled saliva gave similar results after sonic vibration over similar time periods, but counts were reduced if exposure was continued for 45 minutes and a marked reduction was noted after 60 minutes. A few runs to determine the effect of sonic vibration on lactobacillus counts indicated that the counts could be doubled, the optimum exposure time seemed to be between 6 and 15 minutes.

32. Synthesis of Mucinous Polysaccharides by Oral Bacteria. *Marshall L. Snyder and Dorothy D. Johnston, University of Oregon Dental School, Portland, Ore.* Our previous studies have reported the incidence of mucinous polysaccharides synthesis, as evidenced by the formation of large and variable mucinous colonies on 5 per cent sucrose tryptose agar, to be limited to a single bacteriological type, *Strep. salivarious*, and the nature of the polysaccharide formed to be a levan. By the use of penicillin in the medium it is possible to inhibit the growth of this type and detect colonies of bacteria capable of synthesizing dextrans. The size and nature of these colonies never approaches that of the levan forming streptococci. Several hundred specimens of saliva have been cultured on 5 per cent sucrose tryptose agar with and without penicillin (0.1 unit/ml.) under aerobic and anaerobic conditions. After 48 hours incubation half of each plate was sprayed with Gram's iodine for the production of the characteristic reddish-purple reaction with dextrans. Purple-staining colonies were found in 20 to 50 per cent of each group of specimens cultured. There was no appreciable difference in occurrence of these colonies on plates incubated aerobically or anaerobically. All strains producing dextrans were observed to be Gram-negative diplococci. Streptococci of the dextran-forming type (*Strep. s.b.e.* or *Strep. sanguis*) were not encountered. This seemed rather strange in view of their high incidence in bacteremias following extraction of teeth. It is possible that the high dilution of saliva (1-1,000,000) used to obtain adequate isolation partially accounts for this discrepancy. The biochemical and serological properties of these gram-negative diplococci will be reported later.

33. Supporting Evidence That Bacterial Antagonism is a Dental Caries Inhibitory Factor. *Charles A. Scrivener, College of Physicians and Surgeons, School of Dentistry, San Francisco, Calif.* Active caries does not progress under deposits of salivary calculus. Naeslund has presented evidence that specific micro-organisms are responsible for the deposition of salivary calculus. This theory is corroborated by the more recent work of Hewitt and Bartel. A gram-positive bacillus, a gram-negative micrococcus, actinomyces and leptothryx are the organisms utilized by Bartel and found to precipitate

calcium carbonate crystals from a specific culture medium. Ennever, Robinson, and Kitchin stated: "It was noted that none of the caries-free surfaces from which actinomyces were repeatedly isolated became carious during a period of two years." This they interpreted as supporting the concept that actinomycetes are not significant of the cariogenic potential of a plague. The correlation of the above theory with that of bacterial antagonism as a caries-preventive factor, suggested that salivary calculus might be a constant source of microorganisms antagonistic to the growth of bacteria ordinarily associated with active-caries. Our laboratory investigations using a double-pour-plate method of inoculation and oral lactobacilli as the test organism has confirmed this conjecture. Following the observation described above speculation arose concerning the cause of the green dots that frequently appear in positive Snyder's tests. It had been previously assumed that these dots were chromogenic or dense opaque bacterial colonies growing within the culture medium. Now however, we speculated that these dots were merely areas in the agar itself where the color change has been prevented owing to the influence of the organism present in the area. Pure cultures of organisms were obtained from an assortment of these green dots and were found to represent the actinomyces, yeasts, and all the morphological groups of bacteria. When heavy suspensions of these organisms were added to saliva obtained from caries-active persons, allowed to stand for a few hours, and used as the inoculum for Snyder's medium, the characteristic color change did not occur. This result is presumedly caused by the destruction or inactivation of acid-producing organisms in the saliva by the antagonistic influence of the added organisms. Present efforts are concentrated upon the classification of the antagonistic organisms and a study of their growth factors.

VI. Fourth Session: Afternoon, March 22; Abstracts 34-45

34. Fluorescence: A New Tool for the Investigation of Tooth Structures. *George Dickson, A. F. Forziati, and I. C. Schoonover, Dental Research Laboratory, National Bureau of Standards, Washington, D. C.* A new method, which utilizes the natural fluorescence of human teeth, has been developed for the investigation of the structure of enamel and dentin. Fluorescent photomicrographs are obtained from the visible light emitted by very thin tooth sections illuminated with high intensity ultraviolet radiation. These fluorescent photomicrographs provide information on the distribution of the organic content of tooth structures and reveal very clearly certain developmental or growth lines in the tooth. Enamel tufts, lamellae and lines of Schreger stand out brilliantly in the fluorophotomicrographs. High magnification pictures show that the fluorescence of the dentin is concentrated along the dentinal tubules. In general the intensity of the fluorescence appears to be an indication of the relative organic content of the various areas of the tooth. Fluorescent spectroscopy methods are being used to study the differences in the light emitted by various areas of the teeth. The specimens used in this work are ground or polished to a thickness of 25 to 75 microns and mounted on microscope slides by means of sodium silicate. To make the fluorophotomicrographs and the spectrographic recordings the slides are illuminated with ultraviolet radiation from a quartz, 1000 watt, water-cooled, mercury vapor lamp. Suitable glass filters between the lamp and the specimen and, in the case of the photomicrographs, between the specimen and the film absorb the visible light emitted by the lamp and the ultraviolet light transmitted by the specimen.

35. Variations in the X-ray Diffractograms of Dental Enamel of Man and Shark. *Otto R. Trautz, Edward Klein, and H. K. Addelston, New York University College of Dentistry, New York, N. Y.* The diffraction patterns ob-

tained with thin sections of human enamel often show the presence of 2 fiber structures of the apatite crystal aggregates, whose fiber axes intersect at an acute angle and lie in the longitudinal planes of the tooth. This orientation is evident in the earliest stage of mineralization. In the enamel of shark's teeth we find also 2 fiber axes in the longitudinal plane, intersecting at right angles. They run parallel and perpendicular to the surface; they are parallel with the direction of the collagen fibers of the organic matrix in the early stage of its formation. Such enamel studies with x-ray diffraction methods may dictate a new chapter of the comparative histology of dental enamel. With shark's teeth the same differences are observed between the sharp diffraction lines of enamel and the diffuse lines of dentin, as in human teeth; though shark's enamel is produced by mesodermal cells, while human enamel is of ectodermal origin. The sharp lines are a better criterion for a tissue being enamel, than is its solubility in nitric acid. The line profiles of (004) and (410) reflections, measured with a Geiger counter spectrometer, have equal integral breadth, indicating equal crystallite diameters, along the c axis and across. For a mixed sample of powdered human enamel: $a = 9.44_0$ $c = 6.88_8$ $c/a = 0.729_5$ and for a sample of precipitated OH apatite: $a = 9.42_8$ $c = 6.87_9$ $c/a = 0.729_6$ As the unit cell dimensions of shark's enamel apatite are near those of fluorapatite, we conclude that the fluorine is contained in the enamel as fluorapatite.

36. Crystallographic Investigation of Tricalcium Phosphate Hydrate. *Aaron S. Posner, and Samuel R. Stephenson, Dental Research Laboratory, National Bureau of Standards, Washington, D. C.* By use of a hydrothermal bomb technique crystals of chemically precipitated tricalcium phosphate hydrate were grown from about 0.05 microns to about 3 microns in length. By this means it was possible to obtain accurate crystallographic data which was not possible using the smaller crystals. The x-ray diffraction powder patterns of this substance were quite similar to those of hydroxy apatite. On the other hand, the index of refraction, the density, and the atomic $Ca:PO_4$ ratio of tricalcium phosphate hydrate were 1.60 (average), 2.89, and 1.50 respectively. This compares to the values 1.64 (average), 3.13, and 1.67 respectively, for hydroxy apatite. These data suggest that tricalcium phosphate hydrate might be hydroxy apatite with phosphoric acid occluded in the ratio of 0.67 H_3PO_4 to one $Ca_{10}(PO_4)_6(OH)_2$. This investigation sheds some light on the constitution of bone and dentin, whose chemical and crystallographic properties are very similar to those of tricalcium phosphate hydrate, as opposed to the constitution of enamel, whose properties are similar to those of hydroxy apatite. (Published in *J. D. Res.* **31**: 371, 1952.)

37. Further Studies on Decalcification Potentials of Foodstuffs. *B. G. Bibby and H. J. V. Goldberg, Eastman Dental Dispensary, Rochester, N. Y.* The following experiments were designed to test the accuracy of the "decalcification potential":

1. *Animal Studies.*—Hamsters were fed special diets for 90 days; food listed comprised 60 per cent of diet; caries scores were recorded.

FOOD	AVERAGE CARIES SCORE	DECALCIFICATION POTENTIAL
Ritz Cracker	9.6	26
Zweiback Cracker	14.7	35
Cheez-it Cracker	9.9	113
Graham Cracker	47.5	226
Lorna-Doone Cookie	46.9	342

2. *Areas of Retention in Oral Cavity.*—To determine the areas in the mouth retaining food particles after mastication, acrylic splints were fabricated to

cover only the teeth. Preliminary results indicate that of the food particles retained in the mouth after chewing, approximately 75 per cent collect on the teeth and 25 per cent on the mucous membranes.

3. *Liquidity and Solidity of Foods.*—The "decalcification potential" of the same food, both in liquid and solid forms, have been determined.

4. *Modified Techniques.*—An anthrone reagent has been employed to measure the total carbohydrate and individual carbohydrates of various foods and mouth collection. This new technique gives accurate results in a minimum of time. The Nelson colorimetric technique has been adapted to the Somogyi-Shaffer-Hartmann procedure and has given satisfactory results.

38. Effect of Penicillin Dentifrices on Acid Production in Dental Plaques and Saliva. *Finn Brudevold and Roland R. Hawes, Eastman Dental Dispensary, Rochester, N. Y.* Intraoral measurements of the fall in pH in dental plaques following sucrose rinses were taken over a 7 week period in groups of dental hygiene students who used: (a) a powder containing 500 units penicillin per Gm. (11 students) (b) a paste containing 1000 units per Gm. (13 students) (c) a control paste without penicillin (21 students). No reduction was found in the acid producing capacity of the plaques in the penicillin dentifrice groups, but after a 5-week period of brushing the groups differed in acid production in morning saliva collected before toothbrushing and eating. The pH readings after 3 hours incubation with sucrose were 5.5 ± 0.4 for the penicillin powder group, 5.2 ± 0.5 for the penicillin paste group and 4.9 ± 0.2 for the control group. To study the immediate effect of penicillin, mouth rinses (250 units per c.c.) were given followed by sucrose rinses ½, 1, and 2 hours later. The fall in pH in dental plaques was not appreciably changed by the penicillin rinses, but acid production was markedly reduced in salivary specimens. Bacteriologic examinations of saliva showed that the penicillin dentifrices produced a slight reduction in gram-positive organisms, and an increase in resistant forms such as the coli-aerogenes group. The reduction in gram-positive organisms was somewhat more marked in the penicillin powder than in the penicillin paste group. The number of yeasts (*Monilia albicans*) and micrococci did not seem to be affected by the penicillin dentifrices.

39. Synergism Between Saliva and Aureomycin or Terramycin. *Frederick W. Kraus and Vilma Johnson, Tufts College Dental School, Boston, Mass.* The susceptibility of *B. mycoides* to the bactericidal action of aureomycin and terramycin in broth is variable. Therefore, calculated values for the median bactericidal concentrations of the antibiotics were substituted for individual readings of tube dilution "sensitivity" tests. The LD_{50} for aureomycin in broth amounts to 0.041 ± 0.012 μg/ml. With the addition of pooled whole saliva diluted 1:160, the LD_{50} is 0.02 ± 0.007. Saliva alone is not inhibitory to *B. mycoides* under the conditions of the test. With terramycin the magnitude of synergism is similar, since the LD_{50} for the antibiotic in broth (0.090 ± 0.008) is halved by the addition of dilute saliva (0.044 ± 0.011). Synergism with either of these antibiotics is destroyed by preliminary heat treatment of saliva (10 minutes at 98° C.), by sterilization with 1 per cent ethylene oxide, or by filtration through fritted glass. Parotid saliva does not exhibit synergism. However, the absence of synergism with sterile saliva is not sufficient proof of a microbial origin of the synergistic substance. Its presence in 25 individuals indicates that it may be a normal constituent of whole saliva.

40. A Clinical Study of the Effects of a Tooth Paste Containing Sodium Copper Chlorophyllin on Oral Bacteria and Gingival Disease. *Emmett R. Costich and John W. Hein, School of Medicine and Dentistry, Uni-*

versity of Rochester, Rochester, N. Y. One hundred and fourteen women, 19 to 22 years of age, were divided into 2 similar groups on the basis of detailed gingival examinations. At the end of 1 month they were given a second series of control examinations and then assigned to either a control tooth paste or the same tooth paste containing 0.1 per cent sodium copper chlorophyllin. Duration of study was 6 months. Tests or examinations were performed at monthly intervals for the following: gingival pathology, salivary lactobacilli, salivary odor, amount of stains, and amount of oral debris. In addition, for the final 4 month period, streptococcus counts, salivary pH and salivary acid production from glucose were determined at monthly intervals. The material was analyzed by means of I. B. M. punch cards. No significant differences between the control and experimental groups were found for any of the factors tested.

41. The Effect of Penicillin and of Terramycin on the Development of Buccal, Labial, Proximal, and Occlusal Surface Dental Caries in Rats and on Quantitative Counts of Oral Microorganisms. *Robert M. Stephan, M. Rachel Harris, and Robert J. Fitzgerald, National Institute of Dental Research, National Institutes of Health, Bethesda 14, Md.* Penicillin and terramycin were added at levels of 0, 20, 100, and 500 units per gram to a fine particle diet, to determine their effect in preventing the development of smooth surface and occlusal fissure caries in rats. The diet consisted of 32 per cent dry skim milk powder, 64.8 per cent granulated cane sugar, 2 per cent dried whole liver substance powder, 1 per cent cod liver oil (U. S. P.), and 0.2 per cent ethylene diamine tetra acetic acid, which had been shown in previous experiments to give a higher incidence and severity of smooth surface caries than the same diet without the EDTA. Four equal littermate distributed groups of albino rats, 45 animals per group, were used for the penicillin experiment and a similar number in the terramycin experiment, 360 rats in all. The animals were sacrificed and caries scored after 78 days. Results: penicillin experiment: (1) control group: 39 rats developed caries with 241 buccal and lingual surface lesions, 164 proximal surface lesions, and 53 occlusal fissure lesions; (2) 20 units penicillin group: 3 rats carious with 10, 16, and 0 carious lesions respectively; (3) 100 units penicillin group: no caries; (4) 500 units penicillin group: no caries. Terramycin experiment: (1) control group: 36 rats developed caries with 285 buccal and lingual surface carious lesions, 134 proximal surface lesions, and 15 occlusal fissure lesions; (2) 20 units terramycin group: 3 rats carious with 6, 0, and 1 carious lesions respectively; (3) 100 units terramycin group: 1 rat carious with 0, 0, 2 lesions respectively; (4) 500 units group: 2 rats carious with 0, 0, 4 lesions respectively. Different forms of caries resulted from some bacterial action which the antibiotics prevented.

Samples of the oral flora were taken before and during the experiment by rubbing the molar teeth with a sterile cotton swab, and then shaking the swab in 10 ml. of sterile broth. Quantitative dilutions of the broth were then plated out in 4 types of culture media. These were (1) tomato juice agar with 20 μg./c.c. of polymyxin added to suppress gram-negative rods, (2) Rogosa's specific lactobacillus medium, (3) violet red bile agar for growth of coliforms, and (4) crystal violet azide agar with 5 per cent blood for growth of streptococci. Penicillin reduced lactobacillus counts to almost zero in all concentrations tested and there was a gradual reduction of streptococci with increasing penicillin concentration. Terramycin had only a gradual reducing effect on lactobacillus counts, but greatly reduced the number and types of streptococci. There was no marked effect on coliform counts. Since both penicillin and terramycin almost completely inhibited the development of caries in these animals it would appear that on the basis of this study this effect could not be attributed to a reduction of a single type of microorganism.

42. Rate of Enamel Dissolution. *Allan Strålfors, Royal Dental School, Stockholm, Sweden.* Total solubility, i.e., concentration in a saturated solution, and rate of dissolution from the solid into an unsaturated solution should be distinguished. The rate of dissolution, which is of essential importance to the caries problem should theoretically be expressed by the diffusion equation: $\frac{n}{t} = D\ A\ \frac{C-c}{d}$ n, is amount of substance dissolved in time, t; when D is the diffusion coefficient, from surface area A when concentration of saturated layer nearest to solid is C and concentration in outer moving solution is c. Further d is the thickness of stagnant layer of liquid near to solid, in which transport can occur only by diffusion. Experiments were performed on extracted teeth covered all over with wax except for a window of standard size according to Brudevold's technique. Each tooth was put into a tube with 10 ml. buffer solution. The tubes were shaken for one hour in a thermostat at 37° C., then the solutions analyzed for calcium and phosphorus. Series were performed, only one factor varied at a time. Factors investigated were: pH of buffer solution, buffer concentration, shaking rate, solution viscosity, and agar gel cast onto surface in comparison to dissolution without agar gel. Further various organic acids were used as buffer substance, namely, formic, acetic, propionic, butyric, lactic, pyruvic, succinic, malic, tartaric, and citric. The results, statistically analyzed, were in agreement with above mentioned diffusion theory. Thus, lower pH gave higher rate of dissolution since C is higher at lower pH, higher buffer concentration gave higher rate since acid has to diffuse onto enamel as it is used up there, higher shaking rate gave higher rate since d is thereby lowered, higher viscosity gave lower rate since d is increased, and increase of d by means of an agar gel gave lower rate. Different acids gave different rates.

43. The Effect of Sugar on the Diffusion of Hydrogen Ions. *B. K. Forscher and L. S. Fosdick, Chemistry Department, Northwestern University Dental School, Chicago, Ill.* Earlier experimental evidence points to the possibility of a physical role of sugar, when present in high concentrations, in the formation of carious lesions. In those in vitro experiments on artificial production of caries, extensive penetration of hydrogen ion was observed only in the presence of concentrated sugar solutions. This might be explained on the basis of an effect of the sugar on the rate of diffusion of hydrogen ion through a semi-permeable membrane. An increased diffusion rate might account for an increased penetration. In this work a study was made of the rate of diffusion of hydrogen ion through semi-permeable membranes in mediums of varying sugar concentrations. It was found that the rates of diffusion of lactic acid and of hydrogen ion are inversely proportional to the sugar concentration.

44. Fractured Vital Teeth. *Everett C. Claus (Veterans Administration Hospital, Denver, Colo.), Colorado Dental Foundation, Colorado Springs, Colo.* A patient, 51 years of age, had the history of hitting his front teeth at the age of 11, 40 years prior to examination. These teeth had to be removed for prosthetic reasons and, because alveolectomy had to be performed, the 4 incisors were removed in 1 block. These teeth never caused the patient any discomfort and tested vital prior to the removal. Histologic examination revealed a fibrous transformation of the pulp with deposition of new hard substance on the pulpal surface of the dentin. This hard substance showed the characteristic features of dentin ground substance with very few dentinal tubules. Cells are incorporated in certain areas, imitating bone of lamellated type. In some regions, resorption has preceded apposition of this hard substance. The pulp tissue is fibrous and vascular with odontoblast-like cells lining the inner surface of the

hard substance. The fractured ends are covered by the bonelike substance and some bone trabeculae are found between the fracture ends. Nerve bundles and vessels pass from the fractured root tip to the coronal part of the tooth.

45. Psoriasis Gingivae. *Horace A. Brayshaw (Fitzsimons Army Hospital, Denver, Colo.), Colorado Dental Foundation, Colorado Springs, Colo.* Psoriasis is a skin disease of chronic inflammatory nature and unknown etiology. Lesions of the oral mucosa have been reported in the literature, but none on the gingiva. This report presents 3 cases which have been observed and studied clinically and histopathologically. Proliferation of the epithelial ridges, clubbing of the papillae of the lamina propria, thinning of the suprapapillary epithelium, parakeratosis and formation of micro-abscesses are the characteristic microscopic features.

VII. Fifth Session: Evening, March 22; Abstracts 46-59

46. Osmokinesis: An Important Factor in the Dental Caries Process. *G. W. Rapp, Loyola University School of Dentistry, Chicago, Ill.* The concept of osmokinesis, movement of fluids under the influence of an osmotic gradient, is applied to the caries process. It is shown that under the influence of an adequate osmotic gradient, nutritents from the blood circulation of the tooth pulp can be drawn through the tooth, to furnish substrate upon which microorganisms inhabiting the surface or subsurface of tooth enamel can act. This phenomenon will be used to explain several hitherto inexplicable observations concerning the caries process.

47. Phosphate Assimilation and Dental Caries. *Thomas B. Coolidge, Zoller Clinic, University of Chicago, Chicago, Ill.* Measurements have been made of the ability of lactobacilli to assimilate phosphorus and this ability has been compared quantitatively with the ability of the lactobacilli to liberate phosphorus from enamel by producing acid. It is shown that phosphate assimilation by microorganisms may play a role in the production of caries.

48. Further Studies on the Effect of Sodium Monofluorophosphate in Experimental Hamster Caries. *John W. Hein, Frank A. Smith and Dwight E. Gardner, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* Fluorine as Na_2FPO_3 or NaF is equally as effective against hamster caries at the 40 and 20 ppm level (*J. D. Res.* **30**: 466, 1951). It has been shown that Ca ions do not interfere with effect of FPO_3 on in vitro salivary acid production as happens with F (*J. D. Res.* **30**: 466, 1951). The effect of Ca ions on the caries inhibiting ability of NaF and Na_2FPO_3 is the object of this experiment. Seven litters of 31 female hamsters were divided into 4 groups by litter mate distribution. The groups differed by the following drinking solutions: Distilled H_2O; Ca, 21 ppm; F, 20 ppm plus Ca, 21 ppm; F as FPO_3 18 ppm plus Ca, 21 ppm. Caries producing diet and solutions were given *ad libitum* for 74 days. Comparisons were made between the caries scores for each type of molar because each tooth type was at a different stage of development at time fluorides were begun (average of 29 days of age). Results: expressed as caries scores for 1st, 2nd and 3rd molars respectively were: Controls 3.1, 7.1, 2.7; Ca 1.8,* 6.4, 1.4*; Ca + F 1.9, 8.1, 1.4; Ca FPO_3 1.5,* 3.9,* 0.0* (asterisk indicates significant difference from control). Conclusions: Ca Cl_2 gave some protection; saturated CaF_2 solution had little effect except possibly pre-eruptively; Ca^{++} interferes with posteruptive action of Na_2FPO_3.

49. Minimal Caries Activity in Syrian Hamsters. *Paul H. Keyes, Harvard School of Dental Medicine, Harvard University, Boston, Mass.* A carefully in-

bred strain of hamsters has been maintained in a constant environment: a year-round 80° F. temperature and an automatically controlled 14-hour light cycle. Animals are raised in cages with shavings; weaned at 21 days; then transferred to screen bottom cages with the experimental ration; and killed when 120 to 125 days old. Caries activity has been practically negligible in animals subjected to the following dietary manipulations: A. Raised on Purina Lab Chow and transferred to a ration containing sucrose 60 per cent; casein 21 per cent; corn oil 5 per cent; liver concentrate 5 per cent; whole powdered milk 5 per cent; Purina Lab Chow ash 4 per cent; vitamin supplements; celluflour 8 Gm./100 Gm. B. Raised on diet described under A and maintained on same. C. Raised on Purina Lab Chow and transferred to a ration composed of whole wheat flour 30 per cent; sucrose 35 per cent; whole powdered milk 30 per cent; alfalfa 4 per cent; NaCl 1 per cent. D. Raised on ration described under A and transferred to ration described under C. E. Raised on ration described under C and maintained on same. F. Raised on ration described under A and transferred to Ration 100 (Shaw, J. H., *J. D. Res.* 26: 47, 1947). G. Raised on R100 (Shaw, J. H., *J. D. Res.* 26: 47, 1947) and maintained on same.

50. Observations of the Dental Caries Pattern in the Syrian Hamster. *J. F. Volker, University of Alabama School of Dentistry, Birmingham, Ala.* It has been previously reported from these laboratories that the production of dental caries in the molar teeth of the Syrian hamster is facilitated when the experimental animals are permitted access to a modified H.W.C. diet (sucrose completely replacing coarse corn) on the eighth to the twelfth day of age and weaned at the twenty-fourth day. When these animals are sacrificed on the seventieth day of life, caries will be noted in almost all of the molar teeth. Further studies have indicated a distinctive pattern of caries susceptibility for each of the 3 molar teeth. The most susceptible area of the third molar is the distal portion of the occlusal surface. The most susceptible area of the second molar is the buccal groove. The first molar routinely develops circumferential cervical caries. Interproximal caries on the distal surface of the second molar is also a relatively common finding. Because of the consistency of this caries pattern it may be of benefit in determining whether or not various caries inhibitory agents or procedures have a greater or lesser efficiency in the prevention of occlusal, buccal, interproximal, or cervical caries, respectively.

51. Effect of Ingestion of Urea and Other Compounds on Hamster Caries. *David F. Mitchell, University of Minnesota, Minneapolis, Minn.* A previous study showed that the ingestion of dibasic ammonium phosphate and urea reduced hamster caries. The present study tests the hypothesis that the ammonium ion caused this reduction. Eight groups of 30 animals each, 30 days of age, were fed a cariogenic diet for 105 days. Drugs were added to the diet of certain groups. The animals were reduced in number by an epidemic. Group distribution and results follow: I. 12 ♀♀, control, caries score 500 (± 132). II. 30 ♀♀, dibasic ammonium phosphate, 1 per cent of diet, 399 (± 122). III. 12 ♀♀, urea 0.6 per cent, 477 (± 100). IV. 25 ♀♀ urea and dibasic ammonium phosphate 1.6 per cent, 415 (± 119). V. 15 ♂♂ control, 706 ± 171). VI. 19 ♂♂, hydrated dibasic sodium phosphate 2.03 per cent of diet (equiv. w'ts. of drugs of VI and II same) 382 (± 118). VII. 18 ♂♂, urea 0.6 per cent, 581 (± 160). VIII. 26 ♂♂, urea and dibasic sodium phosphate 2.63 per cent, 429 (± 97). p values follow: I-II, .021; I-III, .656; I-IV, .057; V-VI, .001; V-VII, .036; V-VIII, .001. The sodium compound was more effective than the ammonium compound or urea.

52. A Study of the Relative Importance of the Various Salivary Glands on the Development of Experimental Caries in the Syrian Hamster. *C. E. Klapper, University of Alabama, Schools of Medicine and Dentistry, Birmingham, Ala.* Earlier reports from these laboratories have pointed out that in desalivated hamsters the severity and pattern of experimental caries is in sharp contrast to that observed in nondesalivated animals. To investigate further the role of saliva in the production of dental caries, 48 hamsters, 42 to 56 days of age, were placed on an experimental regime. In one-half of the animals both the submaxillary and sublingual glands were removed. Twelve of these animals were maintained on a severe cariogenic diet containing 66 per cent sugar. The others were fed a limited cariogenic diet in which the sugar was replaced by starch. The parotid duct was ligated and severed in the remaining 24 animals. These animals were divided into equal groups. Half of them were placed on the sugar diet, and the remainder were maintained on the starch diet. As was observed in the completely desalivated group, the hamsters eating the sugar diet showed caries in almost all of the molar teeth. The greatest destruction was seen in the first molar and the least in the third molar. On a comparative basis considerably more tooth destruction was observed in the completely desalivated hamsters than in the partially desalivated animals. Only a minimal amount of dental caries resulted in those animals maintained on the starch diet with either the parotid gland or the submaxillary and sublingual glands functioning. These results indicate that the reduced flow of saliva was sufficient to remove the starch from the oral cavity before the slow action of the amylase-forming bacteria had converted the starch to sugar. This suggests that the cleansing action of the saliva was its most important role in relation to dental caries. Additional evidence to support this conclusion was found in comparing the caries pattern of the maxillary molars with the mandibular molars in hamsters fed the sugar diet. In those animals with only the parotid gland functioning there was an appreciable reduction in the amount of tooth destruction on the first molar of the maxilla in contrast to the pattern on the other jaws. These findings signify that the amount plays a more important role than the type of salivary secretion in the prevention of dental caries.

53. Relation of Function to Caries Rate. *H. H. Neumann, W. Lefkowitz, and N. A. Di Salvo, Columbia University and Montefiore Hospital, New York, N. Y.* In an experiment designed to study the effects of vigorous mastication on caries incidence over a period of eighteen months, 102 children received a daily supply of fibrous sugar cane which they chewed under supervision, and 164 controls were again divided in two groups, of which one received daily freshly milled cane juice, and the other neither cane nor juice. The amount of chewing in length of time or in the number of chewing motions was considered of minor importance compared with the force exerted during mastication. Sugar cane was selected, as certain varieties of it have considerable taste appeal to youth and at the same time offer a maximum of hard bulk, which during mastication, does not lose its fibrous structure. The experiments suffered a considerable handicap in the difficulty of obtaining a steady supply of palatable cane and in the generally rather poor dental condition at the outset which interfered in many cases with the ability to chew adequately. The teeth were examined with explorer and mirror and radiographically, and caries was scored by the DMF rate. The results obtained showed 50 per cent less increase of caries in the chewing group compared with the control group. The differences may be accounted for by a purely mechanical detergent action of the fibrous food, or hypothetically by the effect of vigorous function on the enamel structure, causing an increased resistance to caries. Another unknown factor may be at play. (Published in *J. D. Res.* 31: 200, 1952.)

54. Incidence of Dental Caries in Primary Teeth in Children One to Six Years of Age. *Bhim Sen Savara and Theodore Suher, University of Oregon Dental School, Portland, Ore.* Six hundred fifty children attending several nurseries and schools in Portland were examined for dental health. The examination was conducted according to the def index as established by Gruebbel (1944). The number of primary teeth missing due to dental caries was also recorded. The analysis of the data showed that the number of children who had dental caries at 1 year (22.2 per cent) increased to 61.8 per cent at 3 years and to 83.3 per cent at 6 years. The average number of def teeth per child at one and at 2 years was .67 and .83, respectively, which increased suddenly to 2.72 at 3 years. This increase in the average number of def teeth continued; and at 4, 5, and 6 years it was 4.04, 4.76, and 5.13, respectively. The analysis of the data according to the severity of the number of teeth affected revealed that the average figure, or the arithmetic mean, did not give a true evaluation of the picture. Therefore, it was considered imperative to include the median, as well as the range of dispersion, for these age groups to give a better reflection of the central tendency for each age group. The average child had 2.75 decayed teeth and .09 filled teeth at 3 years. This variation was reduced considerably at 5 years, when 3.64 teeth were decayed, and 1.09 were filled. At 6 years the average number of filled teeth increased to 1.60 as compared to 3.40 decayed teeth. The validity of the def index was tested for children under 7 years of age by counting the number of teeth which were missing due to dental caries, the maximum number being .6 per cent at 6 years of age, a figure which is not statistically significant.

55. Placement of Fillings as a Source of Statistical Error of Estimate of Dental Caries Progression Rates. *Julian D. Boyd, John C. Cannon and Robert E. Leighton, Colleges of Medicine and of Dentistry, State University of Iowa, Iowa City, Ia.* Operative treatment of dental decay will alter the subsequent course of caries in the treated area. Furthermore the presence of fillings lessens the numbers of areas susceptible to attack by caries. These outcomes are beneficial insofar as the patient's welfare is concerned, but they will cause notable error of estimate if subjects with fillings are used in experimental studies relating to the control of dental caries through nonoperative means. This report presents still another source of statistical error which will arise when caries extent is being measured through the number of affected (DF) tooth surfaces. By necessity a filling is larger than the zone of caries it is designed to repair, and as a consequence the filled tooth may present more affected surfaces than did the same tooth immediately before it was filled. To gain some quantitative concept of how much statistical error might arise through the placement of fillings, the dental records of 99 children prior to any operative procedure were reviewed and then again immediately after repair had been completed. The difference in the number of DF surfaces in the contrasted records is taken to represent the degree in which the dental fillings involved surfaces not affected directly by caries. The DF score increased in 60 per cent of the subjects. Among those with increased scores, the median increase was 2 tooth surfaces for each mouth: a fourth of those showing increase had 4 or more additional surfaces affected. When the degree of increase was compared with the number of carious surfaces noted initially, the average degree of increase was 25 per cent, and one-fourth of the subjects with increase had 50 per cent or more surfaces added to their DF score because of the placement of fillings. The significance of such statistical errors in the formulation of quantitative conclusions is discussed. (Published in *J. D. Res.* 31: 254, 1952.)

56. An Artificial Mouth for Caries Research. *Ward Pigman, H. C. Elliott, Jr., R. O. Laffre and Leonard Robinson, University of Alabama, Dental School and College of Medicine, Birmingham, Ala.* An apparatus has been devised for carrying out in vitro caries studies. With this apparatus it is possible to approximate some oral conditions better than those reported in similar earlier experiments. For its operation, freshly extracted teeth are fixed in moulding compound and placed in a cylindrical funnel. The tooth surfaces are inoculated with a sample of saliva from caries susceptible mouths. Subsequently a nutrient medium is allowed to flow dropwise on the tooth from a reservoir. In the first work this medium consisted of 0.5 per cent glucose, 0.5 per cent tryptone and 0.5 per cent sodium chloride. Preliminary results of the operation of the apparatus on 10 teeth for an 8 weeks' period are available. Extensive decalcification followed by widespread pitting of the enamel and penetration into the dentin is a routine occurrence. Abraded enamel surfaces are the areas most susceptible to these changes. The experimental procedure has been further modified to include daily toothbrushing. It is hoped that this technique modification will make for localization of plaques and caries formation in the noncleansing tooth surfaces. (To be published in *J. D. Res.*)

57. The Buffering Capacity of Dental Plaques. *B. M. Moore and L. S. Fosdick, Chemistry Department, Northwestern University Dental School, and W. J. Carter, Naval Training Center, Great Lakes, Ill.* The buffering capacity of dental plaques was studied by making simultaneous measurements of pH and total lactate concentration on samples of plaques removed from the teeth before and at five minute intervals after a 50 per cent sucrose mouthwash. From this data, free and neutralized lactic acid concentrations of the plaques were calculated. The buffering capacities of plaques from normal and caries immune individuals were compared.

58. Preliminary Report on the Antimicrobial Attributes of Pine Decoctions. *Frank J. Orland and Frances Broschak, Zoller Memorial Dental Clinic and Department of Bacteriology and Parasitology, University of Chicago, Chicago, Ill.* A report by Shaw (*J. D. Res.* **28**: 495, 1949) and concurrent observations made in the Lobund-Zoller Germfree Animal Dental Caries Project that the presence of wood shavings partially inhibited rat dental caries, prompted this in vitro study. It was found that aqueous extracts of wood shavings curtailed acid production by certain oral bacteria. A study was conducted to learn what types of wood decoctions were inhibitory to oral microorganisms. Specimens of heartwood, bark, leaves, pine needles, and cones were tested. Five gram quantities were steeped in 100 ml. amounts of boiling water for 10 minutes. The filtrate was added to a bacterial medium concentrate. Following pressurized steam sterilization, inoculation and incubation for 48 hours, acidimetric titrations were made with 0.1 N NaOH. Some of the arboreal extracts in the medium reduced acid production by 60 to 90 per cent. Heterofermentative lactobacilli and enterococci were less inhibited than the homofermentative lactobacilli. On the bases of 43 different specimens from 26 tree species assayed bacteriologically, the pines as a group demonstrated the greatest inhibitory attributes in contrast to the hardwood trees. Specifically, *Pinus strobus, Pinus sylvestris,* and *Pinus ponderosa* seemed to provide the most potent antibiotic effect on the oral lactobacilli tested. Biochemical analyses for the active factor in pine wood are under way at present.

59. The Pattern of Mitotic Activity in the Oral Epithelium of Rabbits. *J. L. Henry, J. Meyer, J. P. Weinmann and I. Schour, Departments of Histology and Oral Pathology, University of Illinois College of Dentistry, Chicago, Ill.* The pattern of mitotic activity in the epithelium of the oral mucosa was studied in serial histologic sections obtained from 80 biopsies taken from 54 New Zealand white rabbits 3½ months of age. Twenty-six of these animals were given a single dose of colchicine (1 mg./K) intraperitoneally. The biopsies were taken in 4 groups 6 hours apart on littermate animals. The technique and site of biopsy and the histologic preparation and examination of the specimens were standardized. For each mitosis, the phase, direction of division when possible, and location were determined. A total of 15,914 mitoses was observed and recorded in 12,871 fields containing 1,197,511 cells. Biometric analysis of the findings led to the following conclusions: The mitotic index (the number of cells in mitosis per thousand cells examined) was 5.1 ± 0.29 in normal specimens. At different periods of the day and night the average mitotic index ranged from 3.8 ± 0.18 to 7.2 ± 0.56. Mitotic activity tended to occur in localized areas of the epithelium. Sections from colchicinated rabbits showed that over a period of time the localization traveled over different areas. Sixty per cent of the mitoses in the normal specimens occurred in the basal layer. The remaining 40 per cent were found in the stratum spinosum. The study of planes of cell division in the basal layer indicated that almost half (46.2 per cent) of the daughter cells would immediately become members of the stratum spinosum. A single mitosis was estimated to last 64 minutes. Rate of mitotic activity in the stratum germinativum was 1 per cent every 2.08 hours. One hundred per cent replacement may be completed in 208 hours.

VIII. Sixth Session: Morning, March 23; Abstract: 60-71

60. Mortality of the Syrian Hamster Following Cephalic Irradiation. *Herman Medak and George W. Burnett, Dept. of Dental Research Dental Division, Army Medical Service Graduate School, Washington 12, D. C.* In order to study the effects of local x-ray irradiation upon the oral tissues of the Golden Syrian hamster, it was first necessary to determine their mortality rate with varying dosages and conditions. Animals were irradiated with a General Electric Maxitron Two-Fifty; the physical factors were 250 Kv., 30 Ma., 35 cm std., with a 1 mm. aluminum filter. The bodies of the animals were protected by 2 inches of lead. The central ray of the tube was directed from above to the exposed head of the animal. Five groups of animals, with at least 10 in each group, received a total dosage (in air) of 3000 r, 3500 r, 4200 r, 4800 r and 6000 r, respectively, at a rate of 600 r per day. Four thousand two hundred r or more produced 100 per cent mortality within 12 days after irradiation. Considerable amounts of fluids were lost from the oral cavity within 5 to 8 days, with progressive emaciation and loss of weight. At necropsy, tissue damages which are ordinarily seen following total body irradiation were absent. All animals receiving 3000 r survived for 30 days or longer. Three thousand five hundred r produced 50 per cent mortality in 30 days in the strain of hamsters used in this laboratory. The hamsters receiving lethal doses of irradiation were given daily doses of 500 units of penicillin in 10 ml. of 5 per cent glucose in saline intraperitoneally beginning with the fifth day after irradiation. Such treatment did not prolong the survival time of the irradiated animals, indicating that other factors besides infection with gram positive organisms acted to produce the death of the animals.

61. Uptake of Radiocalcium in Experimental Hyperparathyroidism. *J. R. Jarabak, Department of Anatomy, Northwestern University Dental School,*

Chicago, Ill. Four doses of 150 U.S.P. units of parathyroid hormone were given to an adult rat which had previously been given 20 microcuries of calcium^{45} chloride solution. A control rat of the same weight was given 20 microcurie of calcium^{45}. The parathyroid was injected interperitoneally on alternate days. Two days after the last injection of the hormone, both rats were sacrificed and the skulls were prepared for radioautographs. The uptake of the Ca^{45} was very distinct in the dentin of the molars of the experimental and the control, particularly so in the region of dentoenamel junction. This suggests that the radiocalcium has entered the dentin by the 2 pathways, (a) by way of the circulation, (b) permeation by way of the oral cavity. The diffuseness of calcium^{45} uptake in the alveolar process, particularly in the region of the molar teeth alveoli and the immediate subjacent area suggests the presence of calcium^{45} in the osteoid. There are 2 possible explanations for the presence of calcium^{45} in the osteoid, (a) a circulatory lag which prevents the removal of the calcium from the bone, or, (b) re-uptake in the interval between injections. The uptake of Ca^{45} in the sutures of the cranium indicates that there is very little difference between the experimental and the control. There are a few diffuse areas of uptake in the osteoid in the experimental animal which were not found in the control. The most significant changes were found at the base of the upper first incisors. Calcium uptake in the area of odontogenesis was considerably reduced when compared with the control. What uptake has taken place is not particularly consistent, suggesting Ca^{45} uptake in the interim between injections.

62. Preliminary Studies on the Diffusion of P^{32}, I^{131}, and H^3 Through the Teeth in Rhesus Monkeys. *R. F. Sognnaes, J. H. Shaw, R. Bogoroch, and E. Sweeney, Harvard School of Dental Medicine, Boston, Mass.* The relative rates of diffusion of ions through enamel and dentin was investigated in Rhesus monkeys using both intravenous and external administration of P^{32} (as $Na_3P^{32}O_4$), I^{131} (as NaI^{131}), and H^3 (as HT-O). Controlled external exposure of teeth was obtained by isolation of individual molar crowns in vivo. This was achieved by use of a rubber dam and sealed plastic tubes which contained known quantities of monkey saliva with and without radioactive isotope. Five hours after intravenous administration of either I^{131} or HT-O significant amounts of both isotopes were detected in samples of saliva bathing the molars isolated in vivo. Presence of sucrose in saliva modified the rate at which these isotopes diffused from pulp through enamel. On the other hand, it was not possible to detect P^{32} in saliva during the 5-hour experimental period. Five hours after external exposure of molar crowns to I^{131} both blood and thyroid gland of the animal were radioactive. Uptake of P^{32} from the external could be reduced by the presence of sucrose in the radioactive saliva. The significance of the diffusion of ions through enamel and dentin as well as the radioisotope gradients in these tissues was discussed.

63. Electro Microscopy of Osseous Tissue. *Sol Bernick and Richard Baker, School of Dentistry, University of Southern California, Los Angeles, Calif.* The fine microscopic structures of osseous tissue from various animals has been studied under the electron microscope by the use of ultrathin sections. The procedure for the preparation for ultrathin sections and micrographs of osseous tissue were presented to demonstrate the morphologic orientation of the cells, fibrils, and calcified matrix.

64. Fixation, Demineralization, and Dehydration of Dental Tissues for Electron Microscopy. *John T. Albright, David B. Scott, Vernon M.*

Mosley, and Ralph W. G. Wyckoff, National Institute of Dental Research and National Institute of Arthritis and Metabolic Diseases, Bethesda, Md. Thin sections of enamel, dentin, and pulp have been made from mature and developing animal and human teeth, using a rotary microtome equipped with a thermal expansion adapter and glass knife. Fixatives used have been 4 and 10 per cent formalin, 4 per cent formalin neutralized with NH4OH, Zenker's fluid, 2 per cent aqueous osmic acid, and 2 per cent osmic acid brought to pH 6.5 by addition of a citrate buffer immediately before use. Fixation time has ranged from 15 minutes to 30 days. Acids used for demineralization have been 5 per cent nitric and hydrochloric, and 2 and 5 per cent phosphoric, trichloroacetic and formic. Demineralization has been carried on for periods ranging from 4 to 60 days. A method of freeze-drying has been used for dehydration of tissues prior to embedding. Differences have not been noted between mature enamel and dentin fixed by the various agents, but predentin and pulp have appeared better preserved when fixed in 4 per cent neutral formalin (24 hours or longer), or buffered 2 per cent osmic acid (1-2 hours). The organic constituents of enamel and dentin have appeared better preserved when specimens were demineralized slowly with organic acids at low temperature. The cutting of thin sections of demineralized mature human dentin has been greatly facilitated by freeze-drying, and matrix structure has been best demonstrated in such preparations.

65. Electron Microscopy of the Organic Constituents of Dentin. *David B. Scott, John T. Albright, Reidar F. Sognnaes, and Ralph W. G. Wyckoff, National Institute of Dental Research and National Institute of Arthritis and Metabolic Diseases, Bethesda 14, Md., and Harvard School of Dental Medicine, Boston, Mass.* The electron microscopic structure of the organic components of dentin has been studied in thin sections of human and animal teeth in various stages of development. Sections have been made from specimens fixed in formalin, Zenker's fluid, and osmic acid. Undemineralized developing dentin has been sectioned, as well as mature and developing dentin demineralized in nitric, hydrochloric, phosphoric, trichloroacetic, and formic acids. Specimens have been dehydrated prior to embedding either by standard passage through alcohols, or by freeze-drying. In these various preparations it has been observed that the dentinal fiber is thin-walled and tubular. Evidence of a sheath lining the tubule has not been found, but there are indications that the matrix may be more compact at the immediate periphery of the tubule. The periodic spacings typical of collagen have been clearly seen in the matrix fibrils of both developing and mature dentin.

66. Oral Manifestations of Ionizing Irradiation. II. Effect of 200 Kv X-ray on the Rat Incisor When Administered Locally to the Head. *J. A. English, C. A. Schlack, and F. Ellinger, Naval Medical Research Institute, Bethesda, Md.* Striking changes have been observed in the incisor teeth of rats which were exposed to 1500 r of x-ray locally (200 K. V. P., h. v. 1. 0.82 mm. cu.) Grossly one or both maxillary incisors may be absent, shortened or small and opaque. There may be 1 or 2 additional mandibular incisors. Roentgenographic and histologic findings indicate a similar phenomenon in both jaws, which is essentially a temporary halting of tooth formation. In the maxilla the second-formed portion of the tooth pushes out the first part, which is subsequently lost. In the mandible, the part of the tooth which is formed following irradiation tends to grow lateral to the portion which is first formed, both parts often being retained beyond the 100 day experimental period. Histologically, there is marked disorganization of both the enamel and dentin in the region of inter-

rupted tooth development. The dentin shows inclusion of cells characteristic of lacunae of bone and the dentinal tubules are without a pattern of arrangement. Fibrous tissue is found in the area where tooth formation is incomplete. Three separate experiments have been performed, using 54, 98, and 126 rats, respectively. In the first experiment, 12 animals survived 100 days; 4 showed involvement of maxillary and 7 of the mandibular incisors as described. In the second study, 16 treated rats survived 100 days, and all showed this characteristic dental alteration. In the third study, roentgenograms made 43 days following treatment showed all treated animals had interruption in development of incisor teeth.

67. Penetration of Radiocalcium Through Acrylic Margins. *R. E. Sausen, W. D. Armstrong, and W. J. Simon, University of Minnesota, School of Dentistry, Minneapolis, Minn.* Perfection of marginal adaptation for auto-set acrylic restorations was tested by determining penetration of radiocalcium through the margins. Twenty-four cavities were prepared in sound extracted teeth and subsequently filled with 3 different brands of auto-set acrylic, 12 of them by the compression technique and 12 by the noncompression technique. Six of each 12 restorations were immersed in water immediately, the others allowed to dry. Then all were immersed in $Ca^{45}Cl_2$ solution, sectioned, and their activity determined by radioautographs. No penetration by the $Ca^{45}Cl_2$ molecule was seen within the mass of the acrylic, and in most cases there was no activity on the surface of the acrylic. The radiocalcium penetrated every margin to some extent. The least penetration was seen around those specimens which were placed with the noncompression technique and were allowed to dry for a short time. The most penetration was seen around specimens placed with the compression technique and immediately immersed in water. No appreciable differences were noted between the 3 brands of acrylic used. Control teeth, subjected to the same techniques, demonstrated a surface uptake on the enamel and cementum, but no penetration of the crown except at enamel defects and lamellae. Expansion characteristics of the auto-set acrylics using the non-compression or "brush-in" technique were estimated by recording temperature changes during polymerization. A net expansion was found to result with this technique. The contraction-expansion phenomena of acrylic is similar in trend to that of amalgam, which has also shown less radiocalcium penetration of its margins than have other filling materials.

68. A Study of the Effects of Radiation on the Periodontium of the Dog. Part I: Clinical Study. *R. Earl Robinson, Harold Ray, and Robert Sherman, College of Dentistry and School of Medicine, University of California, San Francisco, Calif.* The effects of radiation on the periodontium of dogs was studied with the expectation that additional information might be obtained on the histopathology of the periodontal lesion from this form of trauma. In addition, certain observed complications of radiation therapy of intraoral lesions suggested the need for fundamental information on radiation damage in this area. Five adult dogs, which had undergone previous experimental surgery were utilized. These dogs lived in the same environment, and were fed the same diet. Intraoral roentgenograms, kodachrome pictures, and thorough clinical examinations of the mouth were made of each dog. Under Nembutal anesthesia, 4 of the dogs were treated with varying doses of localized radiation, and the fifth was reserved as a control. Dog No. 1 was irradiated in the vicinity of the periodontium of the left maxillary canine with 500 R (air) for 3½ seconds. Dog No. 2 was irradiated in approximately the same area with 1000 R (air) for

7 minutes. Dog No. 3 was irradiated in the canine area with 2000 R (air) for 14 minutes and 12 seconds. Dog No. 4 was irradiated in the same vicinity with 3000 R (air) for 21 minutes and 18 seconds. The physical factors were: 100 Kv (kilovolts), 10 Ma (milliamperes), 29 CM.TSD (target skin distance), 2 mm. AL (added filter) giving a HVL (half value layer) of 3.71 mm. AL. Clinical examinations and kodachrome pictures were made at appropriate intervals. On the eighteenth day of the experiment Dogs Nos. 1 and 2 remained clinically normal while Dogs Nos. 3 and 4 showed, in the area of radiation, a slight erythema over the canine and incisor involving the attached gingiva and extending beyond the mucobuccal fold into the mucous membrane. These lesions continued to increase in severity until the forty-second day when they became severely ulcerated. This ulceration increased in severity and a generalized gingivitis throughout the mouth occurred. Concomitant with the local reaction, systemic and behavior changes were noted in these dogs. Dog No. 2 produced a minimal effect. Dog No. 1 showed no change. Intraoral roentgenograms taken 59 days from the day of radiation and compared with the original films, showed no appreciable difference. Pocket formation, recession of the gingiva or any lesion similar to that of the periodontal lesion with the exception of the generalized gingivitis was not observed. The dogs were sacrificed and biopsy blocks were cut 73 days from the date of radiation.

69. Preliminary Report on the Mechanism of Action of Porphyrin Compounds on Lactobacilli. *Wm. David McBride, Naval Dental School, Bethesda, Md. and School of Medicine, University of Minnesota, Minneapolis, Minn.* Stimulation of *Lactobacillus leichmannii* and the inhibition of *Lactobacillus casei* by 5 different porphyrin compounds has been found to be true only under those specified conditions of oxidation of the culture medium. Aerobic conditions obtained by bubbling oxygen through the medium during growth reduced acid production to approximately one-third that found under anaerobic conditions, obtained by completely filling the culture flask. Both *L. leichmannii* and *L. casei* exhibited this sensitivity to oxidation, but to a varied degree. For both organisms the addition of a porphyrin greatly stimulated acid production under aerobic conditions and retarded it under anaerobic conditions. Preliminary results indicate that a vitamin mixture has the same stimulatory effect. Fermentation balance studies have consistently failed to reveal any differences in the ratio of conversion of sugar to lactate under these conditions. Within limits, cell growth has been found to be proportionate to acid production. Hydrogen peroxide has not been demonstrable in these cultures, although more work is being done in this regard. These results are interpreted as indicating that porphyrins directly stimulate or interfere with the synthesis of some enzyme, or is actually included as a co-enzyme or an antivitamin; it is further believed the explanation concerning the alteration of the environment by chlorophyllins, previously advanced by others, is untenable.

70. The Effect of Selective X-ray Irradiation on the Histologic Structure of the Rat Salivary Glands. *William G. Shafer, Indiana University School of Dentistry, Indianapolis, Ind.* Although the effect of x-ray irradiation on the functional activity of the salivary glands is well known, the effect on the histologic structure has not been thoroughly investigated. In the present study, the salivary glands of the rat were selectively irradiated by means of a 2.5 cm. cone. A superficial therapy machine was used with the following radiation factors: 140 Kv, 8 milliamperes, 1 mm. aluminum filter, target distance of 12 inches, and delivery of 133 roentgen units (r) per minute. The amount of

irradiation administered to the animals varied between 480 r and 3500 r, measured in air in a single dose. All animals were sacrificed after 21 days. The irradiated sublingual gland, which is mucous in type, showed little deviation from the normal structure that could be distinguished by either hematoxylin and eosin stain or by phosphotungstic acid—iron hematoxylin, Mallory's stain, or basic fuchsin-hematoxylin—light green stains. The parotid gland, a serous type gland, showed the most pronounced changes, although these alterations were not apparent until a dose of 2400 r was reached. The effects of irradiation on this gland varied markedly even with similar doses in different animals. The essential picture ranged from edema of the gland through more serious changes including pyknosis and chromatolysis of acinar cells, while the cytoplasm often showed features of albuminous degeneration. Some animals showed complete loss of nuclei of acinar cells, although duct cells appeared relatively unaffected. The submaxillary glands, like the sublingual, appeared relatively refractory to irradiation under the conditions of this study.

71. Effect of Aluminum Oxide on the Rabbit Lung. *Harold W. Fullmer and Robert F. Eastman, Loyola University, School of Dentistry, New Orleans, La.* Using 21 rabbits, the effects of aluminum oxide, quartz, dolomite and carbon black on the lung tissue, were observed histologically. The materials were administered according to the method described by E. J. King, J. W. Clegg and V. M. Rae (Thorax **1**: 188, 1946). Aluminum oxide was administered to 12 rabbits; 4 received dolomite, 2 received carbon black, 2 received quartz, and one received nothing. An average of 1500 mgm. of material was administered to each rabbit. The animals were sacrificed after varying lengths of time, the shortest period being 6 weeks; the longest, 17 months. A comparative study of tissue response to the different materials would indicate the following: (1) Quartz caused a marked chronic granulomatous reaction. The granulomatous areas contain a large number of macrophages, foreign body giant cells, and a fibrous tissue proliferation. (2) Aluminum oxide caused a weak chronic granulomatous reaction in which there were many macrophages, a few foreign body giant cells, and a slight fibrous tissue proliferation. (3) The carbon black was walled off by only a few layers of connective tissue cells. There were no giant cells. (4) The reaction to dolomite was similar to that of aluminum oxide.

IX. Seventh Session*: Afternoon, March 23; Abstracts 72-95

72. The Effect of Tobacco Smoke on Epithelial Tissues of Mice as Influenced by Vitamin B Deficiency and Gonadectomy. *Seymour J. Kreshover, School of Dentistry, Medical College of Virginia, Richmond, Va.* An artificial smoking machine was devised so that applications of tobacco smoke could be made to the lips and ears of mice. This procedure was carried out on alternate days and standardized so that the animals received, on each day, a like number of applications of equal volume, exposure time, and interval periods. Sacrifices were at varying periods up to 72 days. Sixty-five male and female mice of pure Swiss and C_{57} strains were treated in this manner. Of this number, 19 were maintained on a normal diet, 19 were rendered deficient in the vitamin B complex, 19 were gonadectomized, and 8 were both castrated and B deficient. When examined grossly, the ears of the normal diet animals showed little evidence of pathosis, whereas, the ears of the B deficient mice were thickened, scarred and, in many instances, excoriated. The gonadectomized group showed no apparent ear abnormalities and there was but slight change noted in the B deficient—castrated mice. No significant gross changes were seen in the lips of any of the

*The Seventh Session was held in two sections.

experimental animals. Histologic examination of the ears showed changes that were most marked in the B deficient group, less so in the normal diet mice, and least in the castrated and B deficient—castrated animals. Among the abnormalities noted were epithelial hyperplasia, hyperkeratosis, parakeratosis, inflammation of the corium, fibrosis and necrosis. Histologic study of the lips showed no uniformity of change in any of the animal groups. In no instance was there any apparent relationship between tissue response and the sex or strain of animal. The ear findings suggest that vitamin B deficiency increases susceptibility to the smoke irritant, whereas, gonadectomy establishes some degree of resistance. The occurrence of variable but minimal lip lesions might be attributed to the action of the tongue and saliva in removing the deposited tars.

73. Water Soluble Mucoproteins of Gingiva. *Milton B. Engel, University of Illinois, College of Dentistry, Chicago, Ill.* The connective tissues of inflamed or swollen gingiva contain appreciably greater amounts of water soluble mucoprotein carbohydrate than do normal tissues. This observation, made from histochemical data, was verified by analytical procedures. Biopsies of 15 healthy and of 38 inflamed gingiva were immediately frozen in liquid nitrogen and fixation was completed by freezing-drying. A small portion of each tissue was prepared for histochemical evaluation using the periodic-acid leuko-fuchsin reagent. Samples of approximately 30 mg. were reserved for quantitative estimation of mucoproteins. To this end the dry tissue was defatted with cold acetone, dried and extracted with cold M/15 phosphate buffer (pH 7). Mucoproteins were precipitated from the perchloric acid filtrate of this extract with phosphotungstic acid. They were determined and expressed as carbohydrate (mannose-galactose) and as tyrosine. In normal tissues the carbohydrate was calculated to be 254 ± 16 mg./100 Gm. and the carbohydrate/tyrosine ratio was 6.5 ± .46. Inflamed tissues gave values of 386 ± 25/100 mg. and 8.4 ± .56, respectively. Statistical tests show the difference to be significant. Progressive alteration of the nature of this mucoprotein in advanced stages of degradation is suggested by a greater relative increase in the carbohydrate fraction. These changes in the soluble mucoproteins associated with gingival inflammation are presumed to be due to enzymic depolymerization or to altered synthesis of connective tissue ground substance.

74. Histochemical Aspects of Abnormal Dentin Formation. *M. S. Burstone, University of Illinois, College of Dentistry, Chicago, Ill.* Recent studies have demonstrated that irradiation of developing dental structures result in abnormalities of odontogenesis. Formation of abnormal dentin especially throughout the pulp chamber is characteristic. Immature mice (2 to 3 days old) were irradiated by subcutaneous administration of soluble radioactive phosphorus P^{32} in dosages of 18 microcuries per gram of body weight. Other mice were irradiated locally by injection of an insoluble radioactive chromic phosphate suspension (50 microcuries) adjacent to the jaws. Seven days following injection the animals were sacrificed, the heads split mid-sagittally and fixed by freezing in liquid nitrogen according to the Altmann-Gersh method. The undecalcified tissues were dehydrated in a vacuum at –32° C., infiltrated and embedded in paraffin, and sectioned at 5 microns. Sections were stained for glycogen and glycoprotein by use of the Hotchkiss reaction. Sections were incubated with amylase in order to remove glycogen and more clearly visualize glycoprotein. Comparable sections were stained with hematoxylin and eosin. Second molar teeth of irradiated mice exhibit abnormal dentin formation in the area of the root portion of the crown and also in the pulp. With the Hotchkiss reaction the irradiated dentin matrix contains a more reactive glycoprotein

complex as contrasted to the controls. The cells associated with formation of this matrix contain cytoplasmic glycoprotein granules. This suggests that irradiation alters the cells of the pulp so that they elaborate highly reactive glycoprotein matrix which calcifies to form abnormal dentin.

75. Growth and Transformation of the Mandibular Joint in the Rat. V. The Effects of Acute Pantothenic Acid From Birth. *Asger M. Frandsen, D.D.S., Hermann Becks, M.D., D.D.S., Marjorie Nelson, Ph.D., and Herbert M. Evans, M.D., Division of Dental Medicine, College of Dentistry and the Institute of Experimental Biology, University of California, San Francisco and Berkeley, Calif.* The mandibular joints of 20 rats deficient in pantothenic acid from birth were studied histologically at different ages. Rats fed a purified diet including pantothenic acid served as controls. The marked progressive changes in the mandibular condyles of the deficient rats were classified into 4 stages according to deficiency symptoms: *Stage I:* The condylar cartilage was decreased in width, the zone of vacuolated cells was absent as well as the major part of the intermediate zone. The invasion zone was narrow with only a few osteoblasts lining the coarse and blunt bone trabeculae. *Stage II:* The decrease in width of the cartilage was still more pronounced, the embryonic zone being extremely narrow. The intermediate zone was calcified. Osteogenesis had ceased and only a few scattered trabeculae remained. *Stage III:* The condylar head showed signs of disintegration. Of the cartilage, only a narrow zone of embryonic cells remained. Bone trabeculae were scarce and part of the lamina compacta missing. Osteophytic bone growth was arising from the neck region of the condyle. *Stage IV:* The condylar head could not be recognized anymore as such. The synovial membranes were necrotic and the continuity of the tissues broken up. Marked osteoclastic activity was noted in the head region while a very active osteophyte formation developed in the neck region and on the superio-lateral surface of the squamosal bone.

76. Occlusal Forces Used During Food Consumption. *A. Yurkstas, Tufts College Dental School, Boston, Mass.* Previous studies here by Brudevold have demonstrated the possibility of measuring vertical forces of a denture wearer during mastication. Modifications of his technique have resulted in greater duplicability of results. The apparatus has been used for study in 2 full dentures, 1 partial lower and a partial upper, on 6 test meals comprising 47 different foods with varying types of preparation. It was found that similar forces were utilized during the course of mastication on the same food regardless of the type of prosthetic appliance worn, and that inter-individual and intra-individual variations usually amounted to less than 10 per cent. The highest forces recorded during this study were 12 to 14 kg. per tooth, and foods commonly found in the diet required an average of 0.3 to 1.8 kg. per tooth for mastication. Raw vegetables required 2 to 3 times the forces needed for chewing the same vegetables after cooking. The total work (kg. strokes) required to render the food acceptable for swallowing was reduced 83 to 96 per cent. Bread crusts and hard rolls required forces as great as 14 kg. per tooth for mastication. Ingestion of liquids during mastication reduced the maximum forces and kg. strokes required for chewing by about one-half.

77. The Effect of the Changing Dentition on Masticatory Function. *F. R. Shiere and R. S. Manly, Department of Oral Pediatrics and the Laboratory for Oral Physiology, Tufts College Dental School, Boston, Mass.* An investigation has been made of 400 children from 6 to 15 years of age to determine the effect of the changing dentition on masticatory function. Masticatory perform-

ance and efficiency, swallowing threshold, food platform area and maximum biting force were determined on both sides of the dentitions. It was possible to predict masticatory efficiency from masticatory performance with considerable accuracy. Masticatory performance with peanuts, P_p, were well correlated with the performances with carrots, P_c, and with the swallowing threshold, U_p. The masticatory efficiency of the children extended over a wide range and averaged about half that of adults who had all teeth present except the third molars. The first and second permanent molars did not improve masticatory function immediately after eruption, but efficiency gradually increased over a period of 3 to 4 years. Efficiency declined after age 10 probably because of the time required for the newly erupted teeth to become functional. Forty-seven per cent of the 400 cases investigated were classed as malocclusions by orthodontic criteria. Only the class 3 cases showed a low area and a corresponding low performance. Other malocclusions were similar to normal cases in their average masticatory function. There was no significant difference in the efficiency of males and females with similar ages and a limited range in area. Area was less well correlated with efficiency (.57) than was found previously among adults. The maximum biting force had little relationship to masticatory efficiency or food platform area. The tendency to prefer the right side in mastication predominated over the tendency to prefer the more efficient side in mastication.

78. Masticatory Function During a Six Months' Period Following Insertion of Dentures. *Paul Vinton and R. S. Manly, Tufts College Dental School, Boston, Mass.* Thirty-eight denture wearers were observed for changes in denture function from the day of insertion, 1 day, 7 days, and 30 days later. Half of the subjects were instructed to use denture powder and half were asked to avoid its use; the 2 groups were balanced as to age, sex, and previous usage of dentures. Twenty-eight of these subjects were also studied 6 months after insertion. Four patients were under 40 years of age, 18 were 40 to 60 years of age, and 16 were over 60. The tests performed were masticatory performance and swallowing threshold (strokes and pulverization) with peanuts, and maximum biting force on right, left and anterior positions of the mouth. During the first month the mean masticatory efficiency remained essentially unchanged for both groups. Those completing the 6 months' test had similar means for the control group and slightly greater for the experimental group. The degree of pulverization at the swallowing point remained the same for both groups, with slightly fewer strokes used by the experimental group at the 6 month test. In each case the patient believed he had made improvement in his ability to chew food. The actual measurement failed to indicate any major change in ability. Possibly subjective adjustment to dentures consists of learning to handle food with comfort and to swallow larger particles of food.

79. Oral Sensory Thresholds of Persons With Natural and Artificial Dentitions. *R. S. Manly, Carl Pfaffman, Donald D. Lathrop, and Joan Keyser, Laboratory for Oral Physiology, Tufts College Dental School, Boston, Mass. and Department of Psychology, Brown University, Providence, R. I.* Judgment of thickness of discs between anterior teeth is similarly acute among denture wearers and persons with a natural dentition. The minimal threshold is 0.18 mm. or 3.3 per cent of discs about 5 mm. thick. The acuteness of hardness judgments by natural and artificial dentition cases is similar when tested on soft rubber discs between anterior teeth. With hard rubber discs the denture wearers are less sensitive. The tactile thresholds on biting surfaces of teeth of natural dentition cases ranged from an average of 1 Gm. on anterior teeth

to 10 Gm. on molar teeth. The median threshold for 8 denture cases was over 125 Gm. at the first premolar position on the denture. Nine out of 10 natural dentition cases were able to detect 2.9 per cent or less $CaCO_3$ in bland pudding, whereas 6 out of 10 denture wearers required over 9 per cent. In general it appears that the sensory discrimination of denture patients is not impaired when the forces and movements involved are of the magnitude used in mastication. When very small forces or movement are encountered as in judgments of texture, the denture patients seem to possess considerable handicap.

80. The Masticatory Efficiency of Partial Denture Cases Among Navy Personnel. *L. F. Abel, Great Lakes Naval Training Center, Great Lakes, Ill., and R. S. Manly, Tufts College Dental School, Boston, Mass.* The purpose of the study was to obtain information concerning oral function of recruits attached to the Great Lakes Naval Training Center before and after they have received necessary prosthetic restorations. Each subject was given function tests before any extractions were performed, before and after insertion of a partial denture, 10 to 14 days later, and 1 month later. The tests include masticatory performance with peanuts and carrots on each side of the dentition, and swallowing threshold with peanuts and carrots on the preferred side. Food platform areas were recorded for both sides and maximum biting force on abutment teeth. Four partials occluding against a natural dentition and 29 partials occluding against a full denture have been tested. Mastication tests with peanuts as test food were well correlated with those with carrots. There was no apparent difference in masticatory efficiencies for partials occluding against a natural or artificial dentition or for free and partials versus tooth-supported partials. The mean masticatory efficiencies were 14 per cent before insertion of the partial, 20 per cent after insertion, 24 per cent after 1 week and 36 per cent at the end of 1 month.

81. A New Method for Observing and Scoring the Development of Dental Disease in Small Living Unanaesthetized Laboratory Animals. *Erling Johansen, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* A new technique is described by which it is possible to study the progressive pattern of development of dental caries and periodontal disease in small living unanaesthetized laboratory animals. By this method it has been possible to study the relationship between food deposits around the teeth, periodontal disease initiation and the development of cavities. It has been found by these studies that dental caries does develop in animals free of periodontal disease while other animals with extensive periodontal lesions have evidenced no dental caries after an experimental period of 132 days. As an example of the special potentialities offered by this technique a study of the effect of a series of unilateral topical applications of sodium fluoride on the molar teeth of Syrian hamsters which had developed small initial carious lesions has been investigated. During 132 days on a caries producing diet the treated animals failed to show either extension of the initial decay or development of new lesions while the control animals showed marked progression of the initial lesions and development of many new lesions. In order to allow simultaneous recordng and quantification of the progressive development of dental caries, periodontal disease and food debris accumulation a special chart based on Keyes method of scoring hamster caries was also developed. (Published in *J. D. Res.* **31**: 361, 1952.)

82. Comparison of Local Anesthetic Solutions in Dental Procedures. *Sidney Epstein, College of Physicians and Surgeons, School of Dentistry, San*

Francisco, Calif. Following the "blind test" method used in previous reported studies, many of the "popular" anesthetic solutions of several manufacturers were clinically evaluated. Solutions were observed for onset of anesthesia, depth of anesthesia, duration, bleeding at site of operation, and side effects. All solutions produced clinically satisfactory anesthesia in terms of factors observed. Individual variabilities of anesthetic agents and vasoconstrictors were observed. None were significant enough to contraindicate use except in selected patient idiosyncrasies.

83. Reinforcement of the Anesthetic Effect of Nitrous Oxide With Trichlorethylene for the Anesthetization of Out-Patients for Dental Surgical Procedures: A Clinical Study. *Harry D. Spangenberg, Jr., College of Dentistry, The Ohio State University, Columbus, Ohio.* Three hundred and fifty consecutive patients were anesthetized for dental procedures with a combination of nitrous oxide, oxygen, and trichlorethylene. Age of patients varied between 2 years and 73 years. Duration of anesthetic varied between less than 1 minute and 45 minutes. Operative procedures were carried out for extraction of teeth, both simple as well as difficult; for the extraction of teeth and trim of process incident to the seating of immediate dentures; for the excision of tumors; and for the enucleation of cysts. There were no untoward reactions, either immediate or remote which were peculiar to the anesthetic agents under study. The degree of relaxation was such that the use of a mouth prop became unessential in a number of procedures. There was no increase of salivation. The patients recovered consciousness as rapidly as when nitrous oxide and oxygen were used alone. There was a notable feeling of well-being during the period of recovery and a lack of discomfort incident to the operative procedure. Patients who normally would be premedicated where nitrous oxide and oxygen were to be used alone, were completely anesthetized by the nitrous oxide, oxygen, trichlorethylene mixture without benefit of premedication.

84. Influence of Fluoride on Calcification. *M. K. Berkut and James C. Andrews, Department of Biological Chemistry, University of North Carolina, Chapel Hill, N. C.* Radioactive rubidium (Rb^{86}) was administered to hamsters on the premise it would act in biological substitution for potassium. Experiments were to detect, indirectly, influences fluoride might exert on potassium within calcifying incisors. Rb^{86} through its half-life (19.8 d) produced measurable radioactivity at termination of experimental periods. The tracer was injected intraperitoneally (1 ml. weekly dose 310 milliosmolar induced radioactive rubidium carbonate) to provide within animals a source of "substitute electrolyte." Data for the low number of animals are not adequate for precise statistical treatment. Nevertheless, incisors of fluoride-treated hamsters (50 ppm F in drinking water, *ad lib.*) extracted after 3 weeks had counting-rates that averaged 30 to 40 per cent below controls. Incisors from normal hamsters beginning their 9th week on fluoride water displayed counting-rates above controls, 39 per cent higher in one case. Increases in radioactivity were not detected in incisor tips and bases after 3 weeks, but pulps of fluoride-treated animals for the same period possessed counting-rates almost twice that of controls. Prolonged fluoride-treatment produced pulps and bases with counting rates higher than controls, 53 and 33 per cent, respectively.

85. Method for Evaluating Cleaning Effectiveness of Dentifrices. *G. Van Huysen and Thomas Boyd, Indiana University School of Dentistry, Indianapolis, Ind.* The teeth of each member of a group of individuals were cleaned and polished. The labial surfaces of the 12 anterior teeth were photographed immediately afterward using Kodachrome film. Each individual

used a dentifrice for 3 weeks. The teeth were again photographed, cleaned, and the subject given another dentifrice for 3 weeks. Thus each subject used each of the test dentifrices 3 weeks. The finished Kodachrome films on each subject were arranged side by side on a uniformly lighted background from left to right in the order of the cleanliness of the teeth portrayed, and the comparative effectiveness of the dentifrices recorded. A standard dentifrice was included in each series of dentifrices tested. Eight dentifrices were examined. Four groups totaling 98 individuals participated. It was possible to show a difference in the cleaning efficacy of different types of dentifrice formulae, and a tooth paste based on insoluble sodium metaphosphate kept the teeth of more individuals cleaner than any other tooth paste tested.

86. Effects of Airborne Fluorides on Children: Preliminary Investigation. *Bhim Sen Savara, Harold J. Noyes, and Theodore Suher, University of Oregon Dental School, Portland, Ore.* Earlier investigations revealed evidence of fluorosis in cattle and vegetation on Sauvie Island. An examination of oral and general health of 68 of 71 children living on Sauvie Island was conducted and augmented with intraoral roentgenograms. Height and weight of each child, along with roentgenograms of the hand and wrist, elbow, knee, and adjacent portions of the long bones also were obtained. There appeared to be no reduction in the incidence of dental caries. The oral tissues showed no pathologic lesions. The enamel of the teeth, which calcified and erupted while the children were exposed to the contaminated atmosphere, demonstrated no macroscopic calcification or hypoplastic defects. Intraoral roentgenographic findings showed that resorption of primary teeth was normal in 75 per cent of the children. Calcification of the permanent teeth was normal in 53.7 per cent, advanced in 33.3 per cent, and retarded in 13 per cent of the children. Twenty-two per cent of the children showed advanced eruption of the teeth, and 10 per cent, retarded eruption. Calcification of the trabeculae, alveolar crest, and the alveolar bone proper did not reveal any striking findings. Microscopic examination of ground sections of 9 carious teeth, extracted during the survey, demonstrated no change or alteration in the pattern of calcification of the teeth. The physique of the children as assessed from Wetzel's grid and Meredith's charts indicated that the majority of the children were in good health. Roentgenograms of the hand and wrist, elbow, and knee examined by two radiologists did not demonstrate any bone changes that could be attributable to fluoride intoxication. The present survey does not reveal any pathologic changes which could directly or indirectly be attributed to fluoride exposure.

87. Lack of Effect of Fluoride on Permeability of Enamel. *W. D. Armstrong and Leon Singer Medical and Dental Schools, University of Minnesota, Minneapolis, Minn.* The mesial or distal surface of freshly extracted human teeth were treated with 2 per cent NaF solution for 5 hours and the other half of the crowns were treated at the same time with 2.7 per cent NaCl solution. The teeth were then soaked overnight in a 6 mg. per cent calcium solution. The crowns were then exposed to ionic solutions of radioisotopes and the teeth were sectioned parallel to the occlusal plane. Radioautographs prepared from these sections allowed an estimate of differences in penetration of the radioisotopes into the enamel on the fluoride treated and untreated sides. No consistent difference in the permeability of enamel to P^{32}, Ca^{45}, I^{131}, and Na^{22} caused by the fluoride treatments has been found.

88. The Effect of Sodium Fluoride Applications on Dental Caries in Adults. *Benton Kutler, and Ralph L. Ireland, Colleges of Medicine and*

Dentistry University of Nebraska, Lincoln, Nebr. One hundred sixty-two medical students had 2 per cent sodium fluoride solution applied topically to the teeth of one side of the mouth. Four thorough applications were made within 2 weeks and at least 2 days allowed between each application. The first application was preceded by a prophylaxis. During these 2 weeks, bite-wing x-ray films were taken and a detailed charting of restorations, missing teeth and carious areas accomplished. The extent of the carious areas was also designated. Each student was numbered as they presented themselves. Those with odd numbers had the 2 right quadrants treated, while those with even numbers were treated on the left half of their mouth. The opposite side in the same mouth therefore served as a control. It was possible through the use of cotton rolls and immediate rinsing to keep the control quandrants free of sodium fluoride solution. One hundred forty-seven of these students were available for re-examination and bite-wing films approximately 1 year later. The mean time elapsed was 13.6 months and the median time being 13 months. The mean age of the students was 24.6 years. The age range was 20 to 46 years. The first and second examinations were made by the same examiner. He had no conscious knowledge as to the side treated, and any possibility of "unintentional knowledge" was eliminated by the fact that the number of right halves treated equaled the number of left halves treated. At the time of re-examination, 294 treated quadrants showed 103 new decayed or filled surfaces. Of the 103 DF surfaces 72 were in 65 new DF teeth and the other 31 were in old DF teeth. The 294 control quadrants at the second examination showed 91 new DF surfaces, 68 of which were in 62 new DF teeth. It is suggested that 4 applications of 2 per cent sodium fluoride within a few weeks is of little or no value in the prevention of dental caries in adults.

89. The Evanston Dental Caries Study: X. The Caries Experience of 6, 7, and 8 Year Old Children with Progressively Increasing Periods of Exposure to Artificially Fluoridated Water. *I. N. Hill, J. R. Blayney and Walter Wolf, Walter G. Zoller Memorial Dental Clinic, University of Chicago, Chicago, Ill.* This report compares the caries experience rates of 4 groups of 6, 7 and 8 year old Evanston children. The first, or base line group (1946), had no exposure to fluoridated water. The second (1948), third (1950), and fourth (1951) groups were exposed to the fluoridated water 12 to 22 months, 35 to 46 months, and 47 to 58 months respectively at the time of examination. The ages of the children in Group four at the time of introduction of sodium fluoride to the Evanston water, ranged from 9 to 55 months. Regardless of age at the time of fluoridation, the crowns of the deciduous teeth of these children had been calcified. However, of the 6 year olds examined in 1951 only the deciduous incisor teeth had erupted when sodium fluoride was placed in the water. The changes in the deciduous dental caries experience rates for the 1951 group was not considered significant when compared to the 1946 base line group. However the dental caries experience rate of the permanent teeth of the same group was reduced by 46.42 per cent when compared to the base line rate in 1946. All data have been evaluated with approved statistical methods. (Published in *J. D. Res.* **31**: 346, 1952.)

90. Deposition of Fluorine in Bones and Teeth of Growing Rats. *I. Zipkin and F. J. McClure, National Institute of Dental Research, National Institutes of Health, PHS, FSA, Bethesda, Md.* White rats were given drinking water containing 10 ppm fluorine as sodium fluoride, starting at age 30 days and continuing for a period of 420 days. At various intervals during this time, representative rats were sacrificed and molar teeth, femurs, and mandibles were

analyzed for fluorine. Fluorine was deposited in the tissues most notably during the growing period. An everdecreasing increment of the ingested fluorine was retained as the animal grew older. Following the period of rapid growth, little or no further deposition of fluorine occurred. In a second study rats were given 10 ppm fluorine in their drinking water starting at various times between 30 and 330 days of age. During all the fluoride periods the rats received equal quantities of fluorine. The analytical results indicate that, as the bones and teeth grow older, even though they had no prior fluorine exposure, they deposit fluorine at a greatly reduced rate. The results of these studies which in general are substantiated by previous similar studies, suggest the following conclusions regarding the metabolism of fluorine: (a) the deposition of fluorine in bones and teeth of rats ingesting 10 ppm of fluorine in drinking water is closely correlated with body growth; (b) a relatively constant level of fluorine is reached by the bones and teeth of rats and is maintained at this level if the fluorine intake remains stabilized; (c) bones and teeth of mature rats retain fluorine at a much slower rate than do similar tissues of growing rats.

91. Lack of Effect of Fluoride Ingestion on Longevity of Mice. *John J. Bittner and W. D. Armstrong, Medical and Dental Schools, University of Minnesota, Minneapolis, Minn.* Four groups of young ZBC strain mice (38 to 42 animals in each group with initial body weights of 18 to 25 grams) were supplied with drinking water containing respectively 0, 5, 10 and 20 ppm of fluoride. The animals were fed a commercial ration reported to contain 22 ppm of fluoride. After 294 days there was no difference in the mortality among the groups. A significant decrease in body weight growth was experienced by the animals receiving water containing 20 ppm of fluoride. The effect produced by 5 and 10 ppm of fluoride in the water was of doubtful significance.

92. The Effect of Fluorosed Enamel on Glucose Fermentation. *G. F. Gurney and G. W. Rapp, Loyola University School of Dentistry, Chicago, Ill.* Previous studies on properties of fluorosed tooth enamel have indicated changes in hardness and acid resistance as compared to normal tooth enamel. This study determined the effect of fluorosed enamel on the process of acid production. It was shown that the solubility of fluoroenamel is adequate to account for an appreciable fluoride ion concentration. The practical significance of this in caries control was pointed out.

93. Reactions of Fluoride with Hydroxylapatite. *Harold G. McCann, National Institute of Dental Research, National Institutes of Health, Bethesda, Md.* Several hydroxylapatites were prepared, the composition of which varied from approximately $Ca_{9.5}(PO_4)_6OH$ to $Ca_{10}(PO_4)_6(OH)_2$. These apatites were allowed to react with sodium fluoride solutions over a range of 2 ppm to 2 per cent (F^-) at temperatures of 20°, 37°, and 100° C. for periods of time varying from 1 day to 4 months. Complete analysis of the solid and liquid phases enabled calculations to be made indicating whether or not hydroxyl had been replaced to form fluorapatite. The type of reaction depended on the hydroxyl content of the apatite. The least basic product appeared to react solely by phosphate exchange. The 100 per cent hydroxylapatite reacted by both hydroxyl and phosphate exchange with a preponderance of the former at low fluoride concentrations and of the latter at high fluoride concentrations. At intermediate hydroxyl concentrations some fluorapatite formed at 100°C. but not at 37°. There appears to be no change in the type of reaction as the solubility product of CaF_2 is exceeded. This was further indicated by the electrolytic conductivity of hydroxylapatite with 0 to 1000 ppm fluoride. A hydroxyl-

apatite containing radioactive P^{32} and Ca^{45} was synthesized and allowed to react with 0 to 1 per cent fluoride. The type and amount of reaction remained about the same up to about 100 ppm fluoride, as indicated by Ca^{45} and P^{32} levels in the liquid, and then gradually the phosphate increased as more CaF_2 formed. The results suggest that the reaction between hydroxylapatite and fluoride depends on 2 essential factors: (1) the concentration of the hydroxylradical, and (2) the concentration of fluoride.

94. Chemical Studies of Periodontal Disease. IX. Estimation of Salivary Putrefaction. *K. A. Piez and L. S. Fosdick, Chemistry Department Northwestern University Dental School, Chicago, Ill.* Amino acids in saliva from patients showing severe periodontitis and from patients free of periodontal disease were determined by quantitative paper chromatography. Both "fresh" and incubated salivas were studied. Nearly all the common amino acids were found. Several other substances giving the ninhydrin test were also present. One was tentatively identified as σ-amino-n-valeric acid; at least 2 volatile amines were found. Highly significant differences were noted between the normal and pathologic salivas. These differences appear to be due to greater amounts of σ-amino-n-valeric acid and volatile amines in the salivas from patients with periodontitis.

95. Epidemiology of Gingivitis in Children. *Maury Massler, Abram Cohen, and Isaac Schour, University of Illinois College of Dentistry, Chicago, Ill., and Board of Public Education, Philadelphia, Pa.*. The prevalence of gingivitis was investigated in 17,079 children living in Philadelphia and 8,882 children living in Chicago suburbs, using the P-M-A Index of assessment. In the children living in Chicago suburbs, the average P-M-A rose from 3.8 gingival units affected per child at the age of 6 years to 9.4 units at the age of 11 years. Gingivitis decreased thereafter and reached a plateau of 5 gingival units affected per child at the age of 16. Philadelphia children of comparable ages showed significantly less gingivitis. The socioeconomic and nutritional factors were similar in both areas and the technique of examination was the same. Since the only apparent difference between the 2 groups was the fact that the children of Philadelphia enjoyed a regular and routine program of oral hygiene, it is assumed that this factor might account for the lower gingivitis experience in the Philadelphia school children. Racial differences in gingivitis experience were investigated in 17,079 white and 6,975 Negro Philadelphia children. Negro children 6 to 11 years of age had a more severe type of gingivitis (especially marginal gingivitis) than did white children of the same age, although the number of children affected was approximately the same in both groups. After 10 years of age, there were no significant differences between Negro and white children. Sex differences in gingivitis experience were examined in a total of 30,031 white and Negro children. No difference in the extent or amount of gingivitis could be demonstrated between boys and girls under 13 years of age. After 13 years, girls clinically showed less gingivitis than did boys. The difference was, however, slight and not statistically significant.

X. Dental Materials Group*; Abstracts 96-104

96. Manipulative Factors Influencing the Physical Properties of Plaster. *J. T. Lindquist and R. W. Phillips Indiana University School of Dentistry, Indianapolis, Ind.* The objective of this study was to determine the

*The meetings of the Dental Materials Group were held simultaneously with the general sessions of the Association on March 22 and 23. In addition to the papers abstracted, two symposia (Self-Curing Denture Base Materials, and Present Day Casting Methods) were presented.

manipulative procedures which would produce optimum physical properties in plaster of Paris. The methods of preparing the mixes were divided into 3 general techniques: spatulation by hand, power-driven mechanical spatulation, and auxiliary use of vacuum. In addition, other factors such as method of adding powder to water, type of mixing bowl, etc., were studied. The physical properties evaluated were compressive strength, microscopic and gross porosity, and density.

97. A Study of the Accuracy of Hydrocolloid Impressions As Employed in Crown and Bridge Technique—Progress Report. *N. E. Hoblit and E. W. Skinner, U. S. Air Force and Northwestern University Dental School, Chicago, Ill.* Two porcelain second molars, approximately ½ oversize, with M.O.D. cavity preparations, have been mounted in a marble block to simulate the position of the teeth for the construction of a hygienic bridge. Platinum markers were placed on the shoulders and occlusal portion of each preparation, and the distance between all of the markers in pairs was noted with a comparator microscope. The report will be concerned with the comparative accuracy of impressions of the model made with (1) reversible hydrocolloid impression material; (2) irreversible material; and (3) a combination of the two.

98. The Microstructure of Dental Amalgam. *D. L. Smith, G. W. Ferguson, and I. C. Schoonover National Bureau of Standards and U. S. Navy Washington, D. C.* Research on dental amalgam is usually conducted with the purpose of finding new methods of manipulation and what effect they have upon the physical properties. Very little work has been done to determine the materials formed when the mercury and alloy react to form an amalgam. A recent study at the National Bureau of Standards has developed a method of determining the microstructure of dental amalgam at room temperature. This was accomplished by an initial polishing on succeedingly finer emery papers followed by a final polishing on satin-covered low-speed laps using three succeedingly finer grits of levigated alumina. Etching was done by two methods: The first was electrolytic, using a high current density for approximately 1 to 2 seconds; the second was by immersion in a solution of approximately 30 per cent nitric acid for 1 to 10 minutes, depending upon the mercury content of the amalgam. Examination of the structure under the microscope revealed that the original alloy particle did not lose its identity, although a higher mercury content resulted in smaller alloy particle remnants.

99. Microstructural Observations of Silver-Tin-Mercury Amalgams. *G. Ryge, Marquette University School of Dentistry, Milwaukee, Wis.* This is a preliminary report of a study of the microstructure of silver-tin alloys and silver-tin amalgams in an attempt to accomplish a better understanding of the setting mechanism. The microstructural changes of an alloy of silver, 73.15 per cent, and tin, 26.85 per cent, annealed for 8, 24 and 72 hours at 425°C will be demonstrated with slides (grain size increasing by time). The dimensional changes and the structure of amalgams with (1) pure silver, (2) pure tin, and (3) silver-tin alloy of different heat treatment and particle size have been registered throughout the setting period. Slides showing the structural changes were shown and discussed. The pictures demonstrate (a) diffusion of free mercury into the alloy particles and (b) growth of amalgam crystals on the polished surface of individual alloy particles—apparently due to diffusion of supersaturated solution of alloy in mercury out of the alloy particle. An attempt is being made at the present to identify the crystals so formed by com-

parison of the reactions occurring in binary and ternary amalgams. The contraction and expansion phenomena observed for the above-mentioned amalgams coincide with the diffusion and crystallization processes described.

100. A Statistical Correlation Between Hardness, Proportional Limit, and Elongation of Cast and Wrought Gold Dental Alloys. *S. H. Bush University of Michigan School of Dentistry, Ann Arbor, Mich.* Statistical correlations between Brinell hardness number and elongation and between proportional limit and elongation have been made, using the manufacturers' values for cast and wrought gold dental alloys. The regression lines determined were of the form $\log Y = mx+b$ where Y is BHN or proportional limit and x is elongation. Separate regressions were found for the soft and very soft cast alloys, all other cast alloys plus anterior bands, and the wrought gold alloys. Values were taken from the charts of 10 manufacturers, and it was found that the relationships given in the report were true for the majority of the alloys within the 3 sigma limits noted in the report. It is felt that such equations will be of assistance to the dentist in predicting some of the mechanical properties of interest to him, and it indicates that these relationships are fundamental to the group of gold alloys.

101. Fins on Metal Partial Dentures. *G. F. Glasson and W. T. Sweeney, U. S. Air Force and National Bureau of Standards, Washington, D. C.* Ideally, to prevent formation of fins on partial denture castings, the thermal expansion of the investments used for making both the cast and the mold should be identical. Recently in an investigation at the National Bureau of Standards it has been observed that a rosin-wax coating applied to the cast to facilitate adhesion of the pattern caused excessive fins on the casting when a certain investment was used. The thermal expansion of specimens of this investment when treated with rosin-wax was greater than the expansion observed on the untreated specimens. Two other investments examined did not show this increase in expansion when treated with the rosin-wax mixture. It was also observed that when the cast and mold had different coefficients of thermal expansion, the most difficulty was encountered from fins if the cast was made of the investment having a higher coefficient of expansion.

102. Principles of Foundational Capacity of Dental Investing Tissues. *E. S. Smyd, Detroit, Mich.* Clinical evidence establishes the truth that a relationship exists between mechanics and biology. It is further known that the investing tissues of the teeth are elastic. With the end in mind of studying the characteristic movements of teeth to torque and the effects of prosthetic structures on such movements, stove bolts were imbedded in masses of Korogel to simulate teeth in investing tissues. Single teeth as well as multiple teeth supporting bridgework may be studied for rotational effects to horizontal forces or eccentric loading. Variables relating to size and lengths of the root portion imbedded in the investing material can be kept at laboratory constancy and with simple though adequate measuring devices a close quantitative analysis may be reached to direction and degree of displacement of teeth to horizontal forces. Mathematics derived from the test support the hypothesis advanced by the writer in a recent article of the J. A. D. A. Fixation of abutments alters the characteristic simple hinge rotation of the individual teeth to produce larger arcs of rotation. Rotation of a structure with fixed abutments occurs upon a center lying in the edentulous bone between the abutments. The abutments move bodily up or down depending upon which side of center they happen to be. Mechanical advantage is gained by the larger orbit of rotation. Foundational stability

varies directly as the radius and is further dependent upon the angle of application of the horizontal force. This work has importance to periodontist and prosthodontist alike.

103. A Study of the Retention of a Complete Upper Denture Base in the Mouth. *R. L. Campbell, E. W. Skinner, and P. Chung, Northwestern University Dental School, Chicago, Ill.* Previous studies on the retention of acrylic denture bases have been made on models. The present study is concerned with repeating the previous work on denture bases in the mouth. The static force required to dislodge the denture in certain areas is in excess of 25 pounds, but in other areas it may be as low as 5 or 6 pounds. The effect of peripheral seal, post dam and relief areas is being studied and the results will be given so far as the research has progressed at the time of presentation.

104. Lubricating Qualities of a Few Dental Materials. *F. H. Freeman, Kerr Manufacturing Company, Detroit, Mich.* This report is of an introductory nature. The study was initiated in an attempt to procure the best possible lubricant to be used on fractional horsepower dental equipment. The work is not comprehensive but offers a method of comparing lubricants and reveals the differences that exist in the lubricating qualities of some of the products available on the commercial market. Most lubricant specifications concern themselves with many properties: viscosity, acidity, color, flash point, carbon residue, foaming tendencies, etc. These factors are important. In addition, the ability of a lubricant to reduce wear and to minimize the power loss in fractional horsepower equipment is also important. This work is concerned with comparisons of these 2 factors for several materials in each of the following classifications: 1. Dental hand piece greases. 2. Sterilizing oils. 3. Automotive oils. 4. Automotive greases. 5. Automotive "lubrication additives."

XI. Papers Read by Title; Abstracts 105-146

105. Effects Upon the Dental Pulp of the Macacus Rhesus of Externally Applied High Frequency Electro-Surgical Currents. *R. Gordon Agnew and William F. Kaiser, College of Dentistry, University of California, San Francisco, Calif.* Surgical currents (vacuum tube circuit—circa 5,000,000 cycles) were applied, at intensities conventionally employed in electrosurgery and with monopolar electrode applicators furnishing either cutting or coagulation effects, to specific areas of incisor, premolar and molar teeth of the Macacus rhesus for varying lengths of time. Histologic examinations, including corresponding controls, made after intervals of 12 and of 4 days. Continuous application to cementum of a molar tooth of loop electrode for ½ minute resulted in widespread pulpal destruction, almost complete disappearance of odontoblasts and profound reticular change. Frequent needle-electrode contact with cementum of adjoining upper first and second incisors during a gingivectomy and gingivoplasty operation (1 minute, 45 seconds duration) designed to simulate careless technique regarding tooth surface contacts, resulted, in the first incisor, only in vascular distension but, in the second incisor, in considerable inflammatory change. One-minute application of ball coagulating-electrode to cervical area of second incisor resulted in extensive localized necrotic change. Needle and loop electrode contacts on bicuspid and molar enamel surfaces showed little effect, in sharp contradistinction to effects of cementum applications.

106. Changes in the Temporomandibular Joint of Rats Caused by Alternations in the Intermaxillary Relationship of the Teeth. *Frank B. Avant, Charles J. Averill and William E. Hahn, Department of Anatomy,*

School of Dentistry, University of Maryland, Baltimore, Md. The purpose of the experiment was to determine if any histologic changes occurred in the temporomandibular joint following disturbances in the intermaxillary relationship of the teeth. Littermate albino rats were divided into 4 groups depending on the operation to be performed: Group 1, extracted upper right molars and first upper left molar; Group 2, ground upper right and left molars and reduced lower incisors approximately 3 millimeters; Group 3, extracted upper right and left molars and reduced lower incisors approximately 3 millimeters; Group 4, extracted upper right molars and reduced lower incisors approximately 3 millimeters. Control animals were included in each group. After 110 days the rats were sacrificed and histologic sections of the joint were made. These sections demonstrated disturbances in the anterior portion of the condyle and comprised replacement fibrosis of the subchondral bone, hemopoietic marrow spaces and cartilage cap; atrophy and rarefraction of condylar cartilage and cessation of bone growth. Morphologic changes were evidenced by a flattening of the condylar head, and by loosening and fraying of the interarticular disk. Histopathology of the component tissues of the walls of the glenoid fossa was limited. The degree of the pathologic disturbances of the joint were in direct relation to the severity of the intermaxillary disturbance.

107. A Study of the Amelogenic Region of the Developing Teeth of Pigs. *James K. Avery, School of Medicine and Dentistry, University of Rochester, Rochester, N. Y.* Further studies of the developing teeth of the pig have been made. It has been noted that the crypt region contacting the forming tooth cusps is an area of great activity in early amelogenesis. The sequence of events in this region has been recorded as follows: (1) The inner and outer enamel epithelium contact each other, (2) dentin formation begins, (3) capillaries from the surrounding crypt contact the stratum intermedium and develop into a sub-ameloblastic plexus over the enamel forming area, (4) enamel matrix formation begins, (5) the cells of the stratum intermedium then rotate their long axis 90 degrees and come to lie in the same direction as the ameloblasts. It thus appears that pathways for calcification of the amelogenic region are established before enamel formation begins. Also the function of the cells of the stratum intermedium in amelogenesis should not be overlooked.

108. Role of Meckel's Cartilage in the Development and Growth of the Rat Mandible. *S. N. Bhaskar, J. P. Weinmann, and I. Schour, Departments of Histology and Oral Pathology, University of Illinois College of Dentistry, Chicago, Ill.* This histologic study on the origin, fate and role of Meckel's cartilage in the growth of the mammalian mandible is based on serial sections of the heads of 84 rats ranging in age from 13 days insemination age to 30 days after birth. Graphic reconstructions of the Meckel's cartilage were made at 15, 16, 17, and 18 days insemination age. Anteroposterior length of Meckel's cartilage and the transverse distance between the right and left Meckel's cartilages were measured at all stages. The right and left Meckel's cartilages differentiate separately at 13 days insemination age in the mesenchyme of the middle of the mandibular process. From here the cartilages grow forward, meet in the midline, and form a process for which the term rostral process is suggested. Prior to the establishment of the squamo-mandibular articulation, the interstitial and appositional growth in the rostral process serves both to lengthen and widen the mandible. After the squamomandibular articulation is formed the Meckel's cartilage is split into 2 parts. The posterior part of the Meckel's cartilage extends from the

middle of the mandibualr process to the head of the malleus, undergoes degeneration, and is replaced by connective tissue. The anterior part of the right and left Meckel's cartilage including the rostral process persists. It continues to grow forward by interstitial and appositional growth at the rostral process, undergoes endochondral ossification, and thus contributes to a great extent to the forward and transverse growth of the mandible. Meckel's cartilage plays a far more important role in the development and growth of the rat mandible than has previously been assumed.

109. Enamel Lamellae and Their Origin. *Charles F. Bodecker, Center Moriches, N. Y.* The occlusal tooth surfaces are regarded as the most vulnerable areas to attack by dental caries, due to the presence of enamel lamellae. Much has been written regarding enamel lamellae, but very little as to their origin. They are generally believed to develop in forming enamel, yet only very few photomicrographs have been published showing even a slight condensation of the organic enamel matrix in embryonic enamel which might be construed as a lamella. Consequently, as these structures are profuse in erupted teeth and almost if not completely absent in forming enamel, another approach to their origin is suggested. It is known generally that enamel lamellae cannot be differentiated from cracks except by decalcification. When ground sections of enamel are decalcified, cracks disappear and lamellae remain as organic, sheetlike structures. Sognnaes (Bodecker, C. F., and associates [*Oral Surg., and Oral Med., and Oral Path.* **4**: 787, 1951]) recently stated his belief, shared by Bodecker, that lamellae are cracks. This concept, however, does not account for the organic structure isolated after decalcification. Bodecker then advanced the concept that the enamel lamella is the residue of dental lymph which is comparable to but not identical with tissue fluid. When trauma causes a crack in a vital tooth, lymph seeps in, while cracks, formed during the preparation of the section for the microscope, remain empty. The dental lymph found in cracks of vital teeth probably remain in a fluid state, facilitating the penetration of microorganisms and their products and may be an important factor in initial dental caries. Only when the tooth is removed and fixed is the lymph coagulated and then isolated as a solid sheet of organic material. It may be advisable to experiment further with metallic salt impregnation of lamellae, so as to close these pathways to invading microorganisms and their products. The concept that lamellae are fluid filled cracks in enamel may explain one manner in which topical fluoridation of teeth reduces dental caries. Under these conditions, fluorides may penetrate to the dentin readily. It appears, further, that the blocking of these cracks may retard the penetration of the tooth destructive factors.

110. Effect of Urea and Other Compounds on Periodontal Scores in the Syrian Hamster. *D. S. Chernausek, School of Dentistry, University of Minnesota, Minneapolis, Minn.* A previously reported study has shown ammoniated products to be associated with a reduced amount of periodontal involvement. The compounds were tested singly and in combination by adding them to the diet. The animals were separated by sex at 30 days of age and were maintained on a soft carbohydrate diet for 105 days. Female animals: Group I control; Group II 1 per cent dibasic ammonium phosphate, $(NH_4)_2HPO_4$; Group III 0.6 per cent urea, and Group IV 1.6 per cent dibasic ammonium phosphate and urea. Male animals: Group I control; Group II 2.03 per cent dibasic sodium phosphate $Na_2HPO_4 \cdot 7H_2O$; Group III 0.6 per cent urea, and Group IV 2.63 per cent dibasic sodium phosphate and urea. Mean periodontal scores. Females: I, 7.1; II, 4.9; III, 5.8; and IV, 4.0.

Males: I, 5.3; II, 2.5; III, 5.6; and IV, 3.5. The results confirm the previous observations. Urea in the concentration present does not appear to be as effective as either the dibasic ammonium—or dibasic sodium phosphate in reducing the periodontal scores. These results paralleled the caries scores, possibly indicating a relationship between this form of periodontal involvement and caries.

111. Some Experimental Effects of the Prolonged Administration of Thiouracil. *H. J. Clausen, Department of Anatomy, University of South Dakota School of Medicine, Vermillion, S. D.* Rats, 21 days old, were given a 0.1 per cent thiouracil solution in their drinking water. Littermate control rats were fed the same food but the thiouracil was omitted from their drinking water. Thiouracil was administered continuously to most of the experimental animals for a period of approximately 625 days. Both the experimental and the control groups were sacrificed at the end of the period and various tissues were removed and prepared for future histologic study. Thyroid adenoma formations of various types developed in the hyperplastic thyroid glands of the experimental rats. Most of these tumors were considered benign but some evidence of vascular invasion was present. However, no evidence of metastases was noted. Approximately 12 per cent of the hyperplastic thyroids presented extensive fibrosis and lymphocytic infiltration which resembled that which is found in struma lymphomatosa. Testicular changes of varying degrees were in evidence. Tubular degeneration and extensive fibrosis of the tunica and blood vessel walls was common. The parathyroid glands of the experimental animals were found to have undergone a change in histologic appearance. The entire gland showed little evidence of a generalized hypertrophy or hyperplasia. There appeared to be an increase in size of the "light chief cells" at the expense of the "dark chief cells." While further investigations are in progress at this time, it appears from a preliminary survey of the material that no structural defects or changes in the enamel or dentin occurred as the result of the prolonged administration of thiouracil.

112. Experimental Studies on the Development of the Mandibular Arch in Amblystoma Punctatum. II. Origin (Preliminary Report). *S. R. Detwiler and B. M. Levy, Departments of Anatomy and Dentistry, Columbia University College of Physicians and Surgeons, New York, N. Y.* In order to verify the neural crest origin of the mandibular arch and its teeth, the following experiments were conducted. Zone 4 (Hörstadius and Sellman) of the head neural crest of *Amblystoma punctatum* in Harrison's stage 15 was removed and grafted into the wound produced by removing the mandibular arch of another embryo in stage 46. At this stage all animals were killed. Serial 10 μ sections through the head were studied. Animals in which the segment of neural crest was removed at stage 15 showed no Meckel's cartilage and no teeth on the operated side. Animals in which Zone 4 neural crest replaced the mandibular arch at stage 30 showed Meckel's cartilage and teeth after reaching stage 46+. All animals were capable of feeding, indicating that function was not impaired.

113. A Corynebacterium Antagonistic to Oral Lactobacilli. *J. Ennever, H. I. Myers, and A. Saeger, School of Dentistry, University of Kansas City, Kansas City, Mo.* From pour plates of 1:1,000 saline diluted saliva in beef heart infusion agar, covered after 96 hours at 35° C. with tomato juice agar pours of *Lactobacillus casei* (ATC-4646) and reincubated 72 hours, a microorganism showing zones of inhibition against the lactobacillus was recovered. The

characteristics of the antagonist demonstrated marked, though not absolute, resemblance to *Corynebacterium enzymicum*. It inhibited lactobacilli isolated from the mouths of 10 subjects but showed no activity against *M. pyogenes var areus, B. subtilis* (ATC-6633B), *E. coli* (ATC-9637), *A. aerogenes* (ATC-8724) and 3 oral yeasts. *C. enzymicum* (ATC-8155, 8156) failed to affect the growth of any of the microorganisms acted upon by the isolated antagonist. As yet, cell free activity has not been obtained.

114. A Quantitative Method for Evaluating the Cavity-Sealing Properties of Restorations. *Joseph Fiasconaro and Harold Sherman, School of Dental and Oral Surgery, Columbia University, New York, N. Y.* A quantitative method has been developed for measuring and recording objectively the cavity sealing properties of dental filling materials. The technique consists of the preparation of a standardized Class V cavity in freshly extracted molars. Restorations are then placed. The root of each tooth is severed, pulp chamber cleaned, and the open portion of the crown individually connected to a brass and rubber tubing assembly. The latter is attached to an air compressor which delivers air through the interior of the tooth crown which is submerged in water. The air pressure is increased until the first bubble appears at the margin of the filling. This technique permits the testing of a given completed restoration under varying physical conditions and at different time intervals. The following standard dental materials were tested and the resultant pressures noted in pounds per square inch: gold foil, 50+, Kyptex, 50+, zinc oxyphosphate cement, 50+, silicate, 38, gold inlay sealed with cement, 45, gold inlay sealed with self-curing acrylic, 20, acrylic inlay sealed with cement, 29. The above figures were accurately reproduced when restorations were carefully placed. However, poorly placed fillings yield considerably lower values. Hence this method may be used to evaluate filling techniques as well as materials.

115. A Quantitative Comparative Evaluation of the Cavity-Sealing Properties of the Self-Curing Resins. *Joseph Fiasconaro and Harold Sherman, School of Dental and Oral Surgery, Columbia University, New York, N. Y.* The method described in the previous abstract was employed to evaluate 5 commercial brands of self-curing acrylics against each other and against the standard restorative materials. The average pressure at which a series of "pressure-matrix" acrylics failed was 5.9 pounds per square inch. The average for the nonpressure or laminated, type of self-curing acrylic restoration was 7.8 pounds per square inch. The latter gave more consistent results. The averages for the commercial brands (non-pressure technique was used) were: Ames 6.0 pounds per square inch, Kadon 5.8 pounds per square inch, Replica 6.6 pounds per square inch, Acralite 6.8 pounds per square inch, Plasto-dent 8.1 pounds per square inch. All of the self-curing resin fillings fail at pressures far below those obtainable from the standard restorative materials which sustain pressures of 38 to more than 50 pounds per square inch. The self-curing resin restorations reach their maximum cavity-sealing value, when immersed in water, in 48 hours. This property increases progressively from 0.6 pounds per square inch at time of filling to 8.2 pounds per square inch in 43.5 hours and remains at 8.2 pounds per square inch. It was found that the cavity-sealing property of self-curing resins is directly proportional to temperature changes. Thus at 32 degrees F. one restoration failed at 4.3 pounds per square inch and improved until at 99 degrees F. it sustained 13.5 pounds per square inch. The adverse effect of radical thermal changes upon cavity seal is most evident in restorations where acrylic is used either as the filling or as cementing medium.

116. Acid Solubility Studies on Enamel and Dentin From Artificially Fluoridated Teeth. *Sidney B. Finn and Charles T. DeMarco, Jr., The Department of Pharmacology School of Medicine and Dentistry, University of Rochester, Rochester, N. Y., and Dental Bureau, New York State Department of Health, Albany, N. Y.* The acid solubility of enamel and dentin samples of exfoliated deciduous teeth from Newburgh children born prior to water fluoridation and after water fluoridation, plus a group of teeth from Rochester, N. Y., children have been compared. Fifty milligram-samples of powdered enamel and dentin were subjected to the action of 20 c.c. of Hac/Na Ac buffer at pH 4 and at 37° C. for 1 hour. Weight losses were as follows: children born in Newburgh 1943 (prior to fluoridation) enamel 13.4 mg., dentin 22.8 mg.; born in Newburgh 1944 (prior to fluoridation) enamel 13.2 mg., dentin 23.9; Rochester, N. Y., children (fluorine free water) enamel 13.9 mg., dentin 23.2 mg.; Newburgh children born in the post fluoridation period, enamel 11.5 mg., dentin 23.4 mg. The differences observed between the enamel from the children born after fluoridation show a significant difference from the children born prior to fluoridation or from the Rochester children when the T test is applied. In all these studies a modification of the standard procedures was used.

117. Morphology of the Cells of the Stellate Reticulum. *Harry E. Frisbie, University of California College of Dentistry, San Francisco, Calif.* Studies were made of stained sections of tooth buds which were dissected from the jaws of dogs, cats, and pigs. These teeth had developed to a stage in which a narrow band of formal enamel capped a wider and more extensive zone of dentin. Fixation was in Carnoy agent or in 10 per cent neutral formol. While it was not found possible to consistently prevent artifact from shrinkage nor from a partial loss of cytoplasm in all specimens, normal relations of groups of cells and at times throughout the full width of the enamel organ were so maintained. In contrast to earlier published descriptions, the cells of the stellate reticulum were found to be large and irregularly shaped and to have the narrow intercellular areas normal to an epithelial tissue. Cells ranged in size up to 50 μ by 75 μ. In these cells only occasional nuclei were seen due to this extreme size. Transitional intermediate cells occurred bordering the external enamel epithelium and also the stratum intermedium. That these 3 layers are probably 1 functional unit is further evidenced by the cellular organization at the sites of enamel organ reduction taking place adjacent to the cuspal tips. Here, the large reticulum cells undergo reduction in size and are blended with the cells of the other 2 strata into a single layer of uniformly small compact cells. During this process the width of the enamel organ is decreased and the cells of the external enamel epithelium are brought closer to the surface of the ameloblastic layer.

118. Airbrasive: Patient Reactions. *Maurice A. Goldberg, School of Dentistry, Georgetown University, Washington, D. C.* During courses in advanced operative procedures including Airbrasive techniques, questionnaires were handed to the patients to establish a record of patient reactions to Airbrasive. Forms were filled out for all patients aged 6 to 74 years, and 1,914 cavity preparations were made. Pain was experienced by 49.7 per cent of all patients. Of those who felt pain 81.7 per cent termed the pain mild and 18.3 per cent thought it was severe. Of the former group 81 per cent thought it was less than that from the burr, 14 per cent thought it was the same and 4 per cent thought it was more. Of the severe group 40 per cent thought it was less than the burr, 40 per cent thought it was the same and 20 per cent thought it was more. Of the total of 1,141 patients, 92.3 per cent prefer to have future

dental treatment done with Airbrasive, 1.3 per cent prefer the burr and 6.4 per cent have no preference. Of those who thought the pain was severe, 73.3 per cent prefer Airbrasive, 13.3 per cent prefer the burr and 13.3 per cent have no preference. In the same form questions were asked with reference to the rubber dam, which were answered by 325 patients, 72 per cent of whom had no previous experience with it. Of the total, 48.3 per cent prefer the use of the rubber dam, 18.4 per cent object to its use and 33.3 per cent have no preference.

119. Histology of the Depigmented Enamel in the Incisors of Vitamin E-Deficient Albino Rats. *Humberto Granados and Henrik Dam, Department of Biochemistry and Nutrition, Polytechnic Institute, Copenhagen, Denmark.* The incisor depigmentation of vitamin E-deficient albino rats is influenced by quantitative and qualitative changes of certain nutrients (*Science* **101**: 250, 1945, Idem **102**: 327, 1945, *Brit. J. Nutrition* **3**: 320, 1949, *Acta pathol. et microbiol. Scandinav.* **27**: 304, 1950), and can be prevented by certain sulfur compounds and redox substances (*J. D. Res.* **29**: 689, 1950, idem **30**: 508, 1951). Chemical studies have shown that in the depigmented incisors, compared with normal control teeth, iron is very greatly diminished both in enamel and dentin, whereas manganese increases markedly in both tissues; magnesium increases slightly in enamel but decreases in dentin; the percentage of calcium and phosphorus remains unchanged in both tissues (*Acta physiol. Scandinav.* **21**: 124, 1950). The histology of the depigmented enamel of vitamin E-deficient albino rats has been studied in ground sections of incisors totally, and partially depigmented (in the latter the structure of both normal and depigmented enamel can be well compared in the same tooth). The vitamin E-free ration used contained 20 per cent casein, 20 per cent cod liver oil and 5 per cent McCollum's Salt Mixture 185. The control diet was supplemented with 20 mg. DL-α-tocopherol acetate per 100 g of the diet. The results have shown: 1. Macroscopically, the depigmented incisors are usually translucent, and have no apparent increase in brittleness. 2. Microscopically, the depigmented enamel also appears translucent, and its total thickness is the same as that of the normally pigmented enamel, no hypoplasia being usually present. 3. Thus, in vitamin E deficiency there occurs a specific inability of the ameloblasts to form and/or deposit the iron-rich pigment without producing enamel hypoplasia. 4. These changes are different from those found in other types of incisor depigmentation, e.g., fluorosis, vitamin A deficiency, rickets, etc., in which cases hypoplasia and chalky opacity of the enamel are usually found. (Leicester: Biochemistry of the Teeth, 1949, and Schour and Massler: Chapter on the Teeth in Farris and Griffith's the Rat in Laboratory Investigation, 1949.)

120. Selenium and Dental Caries. An Investigation Among School Children of Oregon. *Demetrios M. Hadjimarkos, Clara A. Storvick and LeMar F. Remmert. Departments of Foods and Nutrition and Agricultural Chemistry, Oregon State College, Corvallis, Ore.* A study was undertaken to ascertain the effects, beneficial or harmful, of the trace element selenium on the incidence of caries among native born and reared children in the counties of Clatsop and Klamath of Oregon. Epidemiologic studies (*J. D. Res.*, **28**: 415, 1949) showed that the children of Clatsop county showed the highest rates of caries attack (14.4 DMF teeth) and those of Klamath county the lowest rates (9.0 DMF teeth). Certain environmental factors were not responsible for this variability in caries rates. (*Oral Surg. Oral Med., and Oral Path.* **3**: 481, 1950). Altogether, 53 children, equally divided between the 2 counties, served as subjects in this study. A 24-hour urine specimen was collected from each subject. By experiments on laboratory animals it has been shown that the

excretion level of selenium in the urine has a definite relationship to the amount ingested. The analyses revealed that the children of Clatsop county showed higher values of urinary selenium concentrations than those of Klamath county. The mean values were 0.049 p.p.m. for Clatsop and 0.037 p.p.m. for Klamath. The difference between the 2 mean values was tested by the analysis of variance and found to be significant at the 5 per cent level. The findings indicate that there is a direct relationship between the degree of susceptibility to caries and the urinary selenium concentrations. This suggests the possibility that the trace element selenium may render the teeth more susceptible to caries attack. Further studies are needed for a clarification of the selenium-dental caries relationship.

121. Hamster Caries and Periodontal Scores: Effect of Ingestion of Amino Acids. *Edith Z. Helman, University of Minnesota, School of Dentistry, Minneapolis, Minn.* Many recent studies have indicated that the presence of ammonia in the mouth is an important factor in caries control. In view of the fact that saliva has deaminating power (hence, can release ammonia from amino acids) it was desirable to see what effect, if any, increasing the amino acid content of the saliva would have on dental caries. Hamsters, 30 days old, from an inbred colony were separated according to sex and divided into 3 experimental groups of 35 each. The animals of Group A were fed a 15 per cent confectionery sugar diet, those of Group B were fed the same diet with 10 per cent Protolysate (amino acid hydrolysate) added, and those of group C were fed this diet with 5 per cent Protolysate added. Due to a Salmonella epidemic which reduced the number of animals considerably, all the males were sacrificed after 50 days; the females continued on the diets for 100 days. After the 50 day experimental period (males) only Group C showed a significant decrease ($p = .002$) in caries incidence. At the end of 100 days (females) no difference in caries experience between the 3 groups was evident. The periodontal scores of Groups B and C were significantly increased over the control Group A after both 50 and 100 days.

122. The Presence of Chondroitin Sulfuric Acid in Dentin. *W. C. Hess and C. Lee, Georgetown University School of Medicine and Dentistry, Washington, D. C.* Chondroitin sulfuric acid was isolated from noncarious human dentin by the procedure Einbinder and Schubert (*J. Biol. Chem.* **185**: 725, 1950) used for its isolation from bovine trachea. From 1 Gm. of dentin 6.4 mg. of the potassium salt were obtained. The identity of the material was established by nitrogen content and by determination of hexosamine and hexouronic acid. Chromatographic analysis indicated that the hexosamine present was galactosamine. The actual content of chondroitin sulfuric acid in dentin calculated from hexosamine analysis of the dentin itself gave a value of 0.43 per cent, indicating that the method of isolation gave a yield of only 15 per cent. Other methods of isolation gave even poorer yields, attempts are being made to improve the procedure.

123. Progress Report of Air Force Vincent's Stomatitis Research Project 21-23-008. *Warren R. Hester, Carlos F. Schuessler, and Col. T. E. Fisher.* Acute exacerbations of Vincent's stomatitis have been noted to occur during or as a result of long flights at low altitudes as well as short flights at high altitudes. Mitchell and a group of clinicians and research workers first initiated studies on this problem in May 1945. This group disbanded after World War II. We propose (1) to substantiate the dental records data on Air Force flying personnel by examining base installation dental records and (2) to investigate some of the chief possible factors involved in ex-

acerbation of Vincent's stomatitis in flying personnel. A summary listing the number of cases reported by AF Bases was compiled and analyzed by the department of biometrics at the aeromedicine school. Each Dental Surgeon is requested to check the number of cases reported to the School of Aviation Medicine by a questionnaire and compare it with the number listed in the "report of monthly dental services." Discrepancies are reported and at the conclusion of the data-collecting phase, a computation and analysis of the information will be made by the department of biometrics. On the basis of these statistics, the project will be further evaluated for its merits by the School's Medicine Research Council. All cases of Vincent's stomatitis diagnosed by a Dental officer or Medical officer subsequent to August 1949, within the continental limits of the United States were included in this study. Over 5,000 cases are reported in this study.

124. Effect of the Facial Musculature on the Facial Skeleton and Dental Arch Form of the Rat. *Sidney L. Horowitz, and Harry H. Shapiro, College of Physicians and Surgeons and School of Dental and Oral Surgery, Columbia University, New York, N. Y.* Portions of the orbicularis oris and buccinator muscles were removed in young rats to study the influence of these structures upon the bony framework of the face and the form of the dental arches during postnatal growth. Various segments of the musculature of the lips and cheeks were removed. Modifications were noted in the dimensions of the nasal bones and in the maxillary and mandibular diastemata.

125. A Technique for the Histologic Preparation of the Organic Matrix of Enamel. *Valerie Hurst and James Nuckolls, College of Dentistry, University of California, San Francisco, Calif.* Numerous workers have attempted the histologic study of the organic matrix of tooth enamel because of its possible relation to the caries process. The advantage of the method to be reported is believed to lie in its extreme simplicity, obviating the need for complicated apparatus. The external physical support of the matrix is maintained during and after decalcification by means of an agar gel which is permeable to the solutions required in the histological preparation. The internal support is provided by applying a modification of the celloidin acid decalcification method. Human teeth are fixed in 10 per cent neutral formol. The crown is separated from the root; the dentin is then removed. The enamel shell is cut in quarters. Embedding is accomplished by placing the enamel remnants in a 2 per cent solution of agar which, after autoclaving and filtering to give greater optical translucency, is poured over the enamel at about 60° C. Upon hardening, a block encompassing the enamel is cut out of the agar. Infiltration is accomplished by dehydrating the agar block containing the enamel in ethyl alcohol of increasing concentrations followed by absolute ethyl alcohol and ethyl ether. The block is then placed in a 5 per cent solution of nitrocellulose dissolved in absolute alcohol for 2 or 3 days. Acid celloidin for the decalcification process is prepared by adding concentrated nitric acid drop by drop to 10 per cent nitrocellulose to give a final concentration of 5 per cent by volume. The agar block is placed in approximately 50 ml. of acid celloidin, and the process of decalcification is followed by a succession of roentgenograms. Upon reaching the end point of decalcification, the excess acid is removed by transferring the block through several changes of 50 ml. volumes of 10 per cent nitrocellulose spaced at several hour intervals. Infiltration and embedding is completed by placing the block in 25 per cent nitrocellulose and 50 per cent nitrocellulose for two or three days each. Hardening is done by the method of slow air drying in a Stender dish. Sectioning is accomplished on a sliding celloidin microtome at an order

of 6μ. Specimens that have been prepared by this method, a full width of enamel matrix has been recovered which has good orientation and rod detail. Only 2 minor disadvantages of the method have been encountered. It has been found preferable to stain the sections before mounting, inasmuch as the agar does not adhere well to albuminized slides. A second disadvantage is that the agar has a tendency to absorb and retain some dies. A stain employed with particular success has been aniline blue.

126. Evidence of Bacterial Consumption of Organic Nitrogen From Human Enamel. *W. E. Hutton and Lois N. Noda, University of California College of Dentistry, San Francisco, Calif.* Consumption of nitrogen during 8 days of growth at 37° C. of an actinomycete strain (known to produce caries like lesions in hamster molars in vitro) was measured (micro-Kjeldahl), simultaneously with lactic acid production in cultures of variable peptone concentration (0 to 0.2 per cent proteose peptone No. 3, Difco; 5 per cent sucrose; 0.5 per cent K_2HPO_4; tap water) to which washed inocula had been added. A fairly clear-cut relation between peptone nitrogen consumption and lactic acid production was observed. In 8 cultures, peptone was replaced by unfixed enamel stored under refrigeration (powdered to pass a 16 but not a 125 mesh screen) which had been purified by grinding away the surface and inner thirds with stones prior to pulverizing. In these cultures, lactic acid production was determined but not nitrogen consumption because of technical difficulties. Of 6 cultures to which 0.25 Gm. of powdered enamel had been added, 4 produced no more lactic acid than controls from which enamel had been withheld, but of the other 2 cultures, lactic acid was produced, corresponding to a peptone nitrogen consumption of 0.7 and 1.2 mg. respectively. Of 2 cultures to which 0.75 Gm. of enamel had been added, only 1 showed significant lactic acid production corresponding to 1.1 mg. of peptone-N. An attack of the organic-nitrogenous material of enamel preparations of this type is indicated, but the relatively high proportion of negative cultures suggests that bacterial adaptation may be a prerequisite to such an attack.

127. Gerüstmark-like Areas in the Tooth Pulp, Periosteum and in the Epiphysis of Long Bones in Scorbutic Guinea Pigs. *James T. Irving and Paul E. Boyle, School of Dentistry, University of Pennsylvania, Philadelphia, Pa.* Thirty guinea pigs of both sexes weighing from 250 to 600 grams were placed on a scorbutigenic diet (Boyle, Bessey, and Howe, 1940). They were sacrificed from 13 to 34 days later. After autopsy, incisor and molar teeth, knees, and other bones were taken for histologic examination. Either the femur or tibia of all animals showed a gerüstmark in the metaphysis. In a number of the epiphyses closely packed fibroblast-like cells, identical in appearance, were also found. Similar areas were observed under the periosteum. In areas of the tooth pulp no recognizable odontoblasts remained, their position being occupied by fibroblast-like cells. These cells were identical in appearance with those of the gerüstmark in bone. We consider that in scurvy the precursors of the osteoblast remain in the fibroblast form and multiply as such; or possibly mature osteoblasts, when they are unable to deposit bone matrix, divide and give rise to poorly differentiated fibroblast-like cells. The latter sequence of events appears to occur among the odontoblasts of the pulp. The resumption of normal cell morphology and function in the early period of repair of scurvy (other animals) support this hypothesis.

128. Uptake of Radioactive Calcium (Ca^{45}) in Fracture Healing. *J. R. Jarabak, Department of Anatomy, Northwestern University Dental School, Chicago, Ill.* The radius of the right leg was fractured in five rats. One 20

microcurie dose of radiocalcium (Ca^{45}) was given interperitoneally 24 hours before the animal was sacrificed. The animals were sacrificed on the fifth, ninth, fourteenth, thirty-sixth, and seventy-second days, respectively. Radioautographs were made from the fractured and control radii. Regular single emulsion dental x-ray film was used for all autographs. The autographs made of the first animal (5 days after fracture) indicate that there is some uptake in the primary callus, this being of a diffuse nature with the largest amount in the immediate region of the broken fragments. The ninth and fourteenth day autographs indicate a generalized uptake on the region of the callus, indicating that the callus is entering the second stage of repair. The pattern of uptake is in part similar to that of the fifth day in so far as the uptake being greatest in the proximal areas of the fracture. In the thirty-sixth day specimen the generalized diffuseness of Ca^{45} is no longer as distinct; areas or bays of uptake are most prevalent along the outer portion of the callus, indicating an organization of the compacta. The pattern of uptake is similar at the proximal ends of the fracture; however, there is also a definite formation of the medullary canal beginning to take place. In the seventy-second day specimen, the numerous individual areas found in the callus give evidence of coalescence in the periphery of the shaft. There was evidence in this specimen of fusion of the ulna.

129. Masseter Muscle in Condylar Regeneration of the Rat. *J. R. Jarabak and Maclyn Kamins, Department of Anatomy, Northwestern University Dental School, Chicago, Ill.* Gross and histologic studies indicate that condylar regeneration can take place when either one or both of the mandibular condyles are removed. It has also been demonstrated that condylar regeneration takes place in only a limited fashion. The regeneration being limited to a healing of the stump and some attempt being made to duplicate form. In the rat, the lateral pterygoid muscles are vestigeal. In the monkey and in man, these muscles are well developed. The primary function of these muscles in man and in monkey, working unilaterally, is to move the mandible to the opposite side. When these muscles work bilaterally, they protract the mandible. In the rat, the function of the mandible is principally propolinal. Lateral function is very restricted. The strongly developed superficial fibers of the masseter indicate that, they, and not the vestigial lateral pterygoid muscles are instrumental in the forward movement of the mandible in food prehension. Following the resection of the mandibular condyles, the mandible is not distally displaced by the posterior temporal fibers and the posterior belly of the digastric muscles. The anterior position of the mandible which is a position maintained by the superficial diagonal fibers of the masseter makes it possible for condylar regeneration to take place.

130. Bilateral Mandibular Condylar Resection. *Joseph R. Jarabak, and Orion Stuteville, Department of Anatomy, Northwestern University Dental School, Chicago, Ill.* The mandibular condyles of a young monkey were resected in the area of the neck below the attachments of the superior and inferior bellies of the lateral pterygoid muscles. These condyles were removed. Cephalometric x-rays taken 6 months later do not show any appreciable changes in facial growth which might be used as criteria of the effects of this impact on facial growth. The bite was opened in the incisor region, extending from canine to canine. The occlusion in the molar region remained perfect. There remains however a possibility that the deciduous molars were depressed allowing for the opening of the bite in the anterior region. The original articular fossas give evidence of "filling in." A concavity is beginning to develop in the region along the anterior incline of the eminence of the temporal bone. This is the

region in which the "stumps" are now articulating. This indicates that a new pseudo-articular fossa is developing to accommodate the condylar "stumps." Since there is no longer any particular need for the articular fossa previously occupied by the condyles, this fossa is filling in and will become obliterated. The mandibular angle becomes more acute, developing an increased antigonial notch. This is in response to the fact that the masseter must take over the function of the lateral pterygoid muscle. Since the superficial diagonal fibers are attached along the region of the antigonial notch, it is plausible to postulate that this change in the gonial angle is in response to a change in muscular function.

131. Condylar Regeneration in the Rat. *Joseph R. Jarabak, and K. Vehe, Department of Anatomy, Northwestern University, Chicago, Ill.* Regeneration of the mandibular condyle of the rat is analagous in part to fracture healing in long bones. Following mandibular condylar resection, the fossa fills in with a blood clot. On the ninth day, this clot begins to organize into a loose fibrous mass. In the area of the resection, osteoclastic activity is extensive along the jagged corners of the bone. In the sixteenth day specimen, cartilage cells begin to appear. Although they are in the connective tissue above the resected stump, they do not form a distinct area of organization until the twenty-eight day. At this time several bays of cartilage cells unite, forming a crescent over the stump. Below the cartilage cells, numerous areas of osteophytes can be seen. On the thirty-fourth day, the fibrous capsule above the area of the cartilage begins to thicken. The width of this fibrous capsule appears to be the factor instrumental in the amount and length of the regenerated condyle. If the capsule is thin, a very distinct condyle forms, smaller in size than the normal. If on the other hand, the fibrous capsule over the cartilage is thick, the regenerated condyle is short, approximating the ramus, having little or no neck. The articular fossa likewise shows histologic changes. The area above the fibrous connective tissue shows a distinct cartilage proliferation and the formation of osteophytes, indicating that there is a filling in process taking place in the fossa.

132. Clinical and Microscopic Studies in Periodontosis Before and After Treatment. *Maxwell Karshan, Benjamin Tenenbaum and Frank E. Beube, School of Dental and Oral Surgery, Columbia University, New York, N. Y.* In anticipation of the possibility that treatment of periodontosis might throw light on the etiology of this disease, 16 cases have thus far received local dental treatment along with vitamin-calcium-phosphorus therapy. The vitamins administered included vitamins A and D, thiamin, nicotinamide, calcium pantothenate, pyridoxine, folic acid, biotin, and ascorbic acid. The vitamin-calcium-phosphorus therapy was given over a period of from 6 months to 1 year during which time the patients received local treatment. All of these patients had been given an initial complete examination as follows: Full mouth series of radiograms, kodachrome photographs of the gingivae, measurement of the pockets around each tooth, alginate impression for study models, and a biopsy of the interdental gingival papilla between the upper left cuspid and first bicuspid teeth for histologic study. After the period of treatment another complete examination was made. The following conclusions are based on the comparative examinations: In 43 per cent of the cases there was some improvement in the clinical appearance of the gingivas. In these cases the gingiva appeared firm, pink and stippled in many areas. Microscopic examination showed a reduction of cellular infiltration in the subepithelial gingival tissue in 56 per cent of the cases. Suppuration and pocket depth were somewhat reduced in 78 per cent and 70 per cent respectively. Fifty per cent of the cases showed

a decrease in tooth mobility. Radiographic examination seemed to show a slowing down in the loss of alveolar bone in 37 per cent of the cases. Thirty per cent of the patients showed no further migration of the teeth.

133. Variations in the X-Ray Diffraction Patterns of Dental Enamel and their Significance. *Otto R. Trautz, Edward Klein, and H. K. Addelston, New York University College of Dentistry, New York, N. Y.* Variations in positions and intensities of the diffraction lines reflect on the chemical composition of the diffracting crystalline matter (hydroxyapatite, fluoroapatite, etc.). Variations in the sharpness of the diffraction lines indicate limitation of the crystallite size. The increased surface area assumes an important role in the storage of calcium and phosphate and other ions and in their metabolic mobilization. Variations in the pattern due to preferred orientation of the crystallites to form fiber structures. These structures are correlated with the orientation of protein fibrils in the developing tissue. There is a variety of such fiber structures in the enamels of different animals; ectodermal and mesodermal enamel.

134. Development of Enamel. *Thorvald Kvam, University of California, College of Dentistry, San Francisco, Calif.* Most vertebrates have their teeth covered with a well-calcified stratum. In fish and amphibians, this part of the crown is not covered with ectodermal enamel, but the dentin becomes more densely calcified than the remainder of the tooth. In reptilian and mammalian teeth, there is an enamel layer deposited outside the dentin by ectodermal ameloblasts. All previous investigations of the development of ectodermal enamel have been studied on man or on species in which the enamel development deviates very little from man. In order to better understand enamel development in high mammals, it would seem that a study of the transitional phases of the evolution of enamel development might be pertinent. The purpose of these investigations is to study the enamel development in reptiles and especially higher ones, such as the alligator, and the lower order of mammals, as the bat. The material consists of sections of entire jaws and of isolated toothgerms. The organic matrix of erupted teeth have also been studied in decalcified sections. In the developing reptilian enamel, Tomes' processes do not seem to occur, but the ameloblastic cells join the enamel matrix to form an even line between the two. Stellate reticulum is found, but the relative amount varies in the different species. The pattern of the enamel matrix is not as regular as in that of mammals. Certain structural arrangements give the appearance of Retzius' lines. Structures which appear as bundles of fibers extend from the dentinoenamel junction to the ameloblasts. These bundles are more definite nearer the ameloblasts and become less distinct toward the dentinoenamel junction.

135. The Embryology of the Rat Molar: Preliminary Report. *William Lefkowitz, Charles F. Bodecker and Dorothy Mardfin, Montefiore Hospital, New York, N. Y.* The use of the first molar of the rat as an explant requires the knowledge of development and growth at definite periods of time. This report is concerned with the development of the first molar of the rat. The Rockland strain of rats which has a period of gestation of 22 days was used. Using the Long and Evans method for the determination of the estrus cycle, it was possible to secure rat embryos of known age. Sections were cut of embryo heads of age 14 days in utero to 5 days post partum. All specimens were taken at 24 hour intervals. The general lamina appears mesial to the area of the first molar and arises from the basal layer of the oral mucosa. It invaginates into the dermis and proceeds posteriorly in the form of an epithelial tube containing an outer enamel epithelium, stellate reticulum, stratum intermedium and inner

enamel epithelium. Histodifferentiation occurs immediately. All the peripheral epithelial cells in the general lamina are columnar. The first molar tooth germ arises as a widening of the general lamina which continues posteriorly to give off the 2nd and 3rd molars. The general lamina maintains its contact with the oral mucosa only at its original site. Differentiation of odontoblasts occurs at the twentieth day in utero. Apparent shrinkage of the dental papilla must be further examined as a normal process rather than an artifact. Further studies of the development of nerves in the dental papilla are in progress. The investigation will continue to study rat embryos at earlier stages to offer a progressive picture of the complete development of the rat molar.

136. The Occurence of Smooth and Rough Forms of Streptococcus Salivarius After the Use of Penicillin Tooth Powder. *V. F. Lisanti and F. R. Shiere, Tufts College Dental School, Boston, Mass.* Streptococcus salivarius counts were carried out on 62 school children. Twenty-nine children served as controls using a base tooth powder with no antibiotic. Thirty-three children were in the experimental group using the same powder with 500 units of penicillin per gram. Duplicate poured plates were counted for the occurrence of rough and smooth forms at 10^5 dilution after 72 hours incubation. In the experimental group those without caries showed a median of 96.5 per cent rough colonies, 4.5 per cent smooth, the control 83 per cent versus 17 per cent, respectively. Those with caries in the experimental group, 85.5 per cent rough colonies, 9.5 per cent smooth, the control group with caries, 79.5 per cent rough and 24.5 per cent smooth. Without regard for caries lesions, the experimental group demonstrated a median of 91 per cent rough and 7 per cent smooth, the control group, 81.2 per cent rough and 20.5 per cent smooth. These data indicate that the penicillin tooth powder after three years use may have altered the types and occurrence of *Streptococcus salivarius.*

137. Resorption Potential of Human Permanent Teeth. *Maury Massler and Anthony J. Malone, College of Dentistry, University of Illinois, Chicago, Ill.* The frequency and degree of root resorption were studied in 700 sets of unselected full-mouth roentgenograms of adults. It was found that 86 per cent of the teeth examined (in 100 per cent of the persons examined) showed evidence of some apical root resorption. There was no evidence of resorption in only 1.6 per cent of the teeth. In 12 per cent of the teeth, resorption of root-ends was considered to be questionable. The cause of root-end resorption was evident in only 5 per cent of the teeth (periapical infections and root canal therapy). In 81 per cent of the teeth examined, no reason for the resorption was evident. The idiopathic resorptions were mild (apex of root slightly blunted) in 71 per cent; moderate (2 to 4 mm. of root resorbed) in 9 per cent; severe (4 mm. to ¼ of root resorbed) in 0.3 per cent; and very severe (more than ¼ of root resorbed) in 0.11 per cent of the teeth examined roentgenographically. The anterior teeth were found to be the most susceptible to apical resorptions while the second molars were least often and less severely affected. The resorption of permanent tooth roots can be greatly accelerated by local factors (orthodontic tooth movement, periapical irritations). This tendency varied considerably in different persons and at different ages. It was concluded that a definite resorptive potential is resident in (or about) the roots of permanent teeth of all persons. While the resorptive potential of the permanent tooth roots is much less than the roots of primary teeth, it is always present to some degree and should be carefully considered prior to orthodontic treatment.

138. The Micro Antimony Electrode. *B. M. Moore and L. S. Fosdick, Chemistry Department, Northwestern University Dental School, Chicago, Ill.*

Micro antimony electrodes are ordinarily very difficult to prepare because of the crystalline nature and brittleness of the antimony. Antimony plated on fine platinum wires seems to provide a satisfactory electrode, except that they are not very durable. When the plated antimony electrode is bent the antimony chips from the platinum wire, thus spoiling the electrode. In order to stiffen the micro electrodes so that a minimum of bending occurs it was decided to use glass as a supporting structure. A fine point was drawn on the end of a glass tube in such a manner that a 40-gauge platinum wire could be sealed into it so that the end of the platinum was brought to the surface of the glass point. The tip was silvered by the usual procedures, after which a coat of antimony was deposited over the silver by distillation in vacuum. In this way a bright antimony deposit was dispersed over the glass point and platinum wire and yielded very satisfactory results for the measurement of pH when used in connection with the vacuum tube potentiometer. The electrodes are quite stable.

139. The Use of a New Electronic Device as an Aid in Oral Diagnosis. *George T. Perkins, and J. Emerson Colby, Dental Research Department, Percy Jones Army Hospital, Battle Creek, Mich.* A preliminary study to determine temperature of the teeth and supporting structures is being made by means of a new electronic device in an attempt to establish a normal temperature distribution in the oral cavity. These measurements are made by the application of thermistors, highly sensitive devices (0.0005° C.), used for recording thermal changes. These thermal changes are then converted into electrical phenomena in terms of resistance by standard electronic procedure. The results obtained will be recorded from temperature-resistance calibration curves and oscillographic recordings on film and tape. Thus, accurate recordings of our experimental data will be kept. Our results will be based on correlating the temperature findings on clinically known pathologic conditions with the histologic reports of the Armed Forces Institute of Pathology and the normal base-line. In this way, we hope to measure and study the possible existence of temperature differences during the different stages of pathology and thus, establish significance for varying degrees of temperature. Preliminary findings in acute pathologic conditions thus far indicate a definite rise in temperature of the affected tooth.

140. Tooth Pulp and Dentin: Respiration. *P. Pincus, Departments of Dental Research and Biochemistry, University of Melbourne, Australia.* Human tooth pulp is found to respire in the Warburg apparatus, utilizing glucose with release of CO_2. In dentin and in pulp both oxidase and succinic dehydrogenase are present, the latter being inactivated by malonate. The succinic dehydrogenase of dentin is inactivated by fluoride at a concentration of about 1 part in 100,000; in these latter experiments, enzyme activity is demonstrated by reduction of methylene blue. The effect of other interfering substances is being investigated. Fluoride is found in some silicate cements.

141. Epithelial Downgrowth on Molar Roots of Hamsters: Effect of Methyl Testosterone. *Martin A. Rushton, Department of Dental Medicine, Guy's Hospital, University of London, England.* Male golden hamster weanlings were maintained up to 9 months of age on a high sugar powdered diet (controls) and the same diet with the addition of 8 milligrams methyl testosterone per 100 grams diet. Serial coronal sections were cut in paraffin, 6 heads from each group, and measurements recorded from the amelo-cemental junction to the deepest point of the epithelial downgrowth in contact with the cementum at fixed anatomical points throughout the molar series. (Basic diet and method

of recording as in *Brit. D. J.* **90**: 87, 1951.) Experimental group remained healthy but fur became stary and final weight was less than controls. The mean extent of epithelial downgrowth was reduced by one-third compared with controls in both jaws but the distribution pattern was similar. There was no difference in the keratinisation of the oral mucosa. The thickness of the dentin of the unerupted part of the lower incisors was less in the experimental group by about half at 9 months.

142. Preliminary Statistical and Morphological Analysis of the So-called Transitional Cell Carcinoma. *Henry H. Scofield, and Joseph L. Bernier, Armed Forces Institute of Pathology, Washington, D. C.* In view of existing confusion regarding the nature and origin of transitional cell carcinoma or so-called lymphoepithelioma, a statistical and morphological analysis of a selected group of these neoplasms was undertaken on material in the files of the Registry of Oral Pathology. Biopsy, surgical and autopsy material from 265 nasopharyngeal and oropharyngeal carcinomas were studied, including both the squamous cell and the transitional cell varieties. In the histologic appraisal, particular emphasis was placed upon the transitional cell carcinomas. To date review of 189 cases has been completed and follow-up information obtained. The following data are referable to this group of cases: Transitional cell carcinoma occurred in 106 of the patients and squamous cell carcinoma in 83. The median age among the former was 38.10 years and among the latter, 56.2. Thus in this series, transitional cell carcinoma occurred in a much younger age group than the squamous cell carcinoma. The youngest patient was 6 years of age and the oldest 75. Of the 106 cases of transitional cell carcinoma, 72 (68 per cent) occurred in the nasopharynx; 23 (22 per cent) in the oropharynx, and 11 (10 per cent) in either the nasal chamber or paranasal sinuses. Of the 83 squamous cell carcinomas, 18 (22 per cent) arose in the nasopharynx; 61 (73 per cent) in the oropharynx, and 4 (5 per cent) in the nasal chamber or paranasal sinuses. The most common initial symptoms of the transitional cell group were: swelling in the neck, 45 per cent; sore throat and dysphagia, 42 per cent; and symptoms referable to the ear, 40 per cent. In the squamous cell group sore throat and dysphagia were the most common symptoms, occurring in 88 per cent of the patients. Symptoms which occurred significantly more often in the transitional cell type than in the squamous cell type were nasal obstruction and epistaxis, swelling in the neck and ocular and aural disturbances. In 92 cases of both transitional and squamous cell carcinoma in which autopsy material was available, there was no apparent difference in the incidence of metastases to specific organs. In 3 cases of transitional cell carcinoma, the spleen, in addition to other organs, was involved. In cases followed 3 or more years there was no apparent difference in survival rates in patients with either type of carcinoma. Ten of 16 patients with transitional cell carcinoma who lived 3 or more years, had no demonstrable recurrence or metastasis at the time of last follow-up.

143. Growth at the Fronto-Nasal Suture of the Rabbit: A Gross and Serial Roentgenographic Study by Means of Metallic Implants. *Abbe J. Selman and Bernard G. Sarnat, University of Illinois College of Dentistry, Department of Oral and Maxillofacial Surgery, Chicago, Ill.* The contribution to the growth of the face of the fronto-nasal suture was studied in 25 growing female New Zealand albino rabbits. The method employed was that of surgically exposing the suture and inserting amalgam into a prepared cavity in the frontal and nasal bones. The distances between the paired implants and the implant to the suture were measured. At the end of the experimental period (5 days to 12

weeks), the animals were sacrificed, the skulls dissected, and the same measurements repeated. In addition, serial cephalometric roentgenograms were taken at 14-day intervals. From these roentgenograms the increased separation of the radio-opaque amalgam pegs could be determined for specific periods. The findings revealed that 1) there was separation of the pegs at the fronto-nasal suture, 2) the rate was inverse to the increase in age, 3) the nasal bone contributed approximately 65 per cent and the frontal bone contributed approximately 35 per cent at the fronto-nasal suture to the separation of the pegs.

144. Clinical Evaluation of a Self-Curing Resin. *Harold Sherman, Joseph Fiasconaro, and Edward A. Cain, Jr., School of Dental and Oral Surgery, Columbia University, New York, N. Y.* Various commercial products were used as they appeared on the market. After a 2-year period, it was found that, out of four products, only one had been used in sufficient cases for a proper clinical evaluation. All restorations were placed by the pressure-matrix method—under rubber dam conditions. A total of 83 cases were evaluated. There were 54 Class III and 29 Class V preparations. Nine cases were examined after 24 months, 45 cases after 18 months and 29 after 12 months. Each case was checked, simultaneously by the 3 authors, for margins, color, hardness, porosity, contour, finish, abrasion, and sensitivity. The data were tested for statistical significance by Neal W. Chilton. It was found that marginal accuracy deteriorates as clinical time increases. Thus at 21 months, 86 per cent appear clinically satisfactory, at 18 months only 44 per cent and at 24 months 33 per cent. The greatest loss in color value is noted clinically during the first 12 months, at which time only 31 per cent of the restorations exhibit satisfactory color retention. While 94 per cent showed acceptable hardness, feather-edge margins failed in all cases. The properties and clinical behavior of the various marketed resins differ considerably. The authors wish to caution the profession that self-curing resin fillings are still in a trial period.

145. Condylar Plating for the Reduction of Condylar Fracture. *Orion Stuteville, and Joseph R. Jarabak, Department of Anatomy, Northwestern University Dental School, Chicago, Ill.* The right mandibular condyle was resected in the region of the neck below the attachments of the lateral pterygoid muscles. The articular capsule was opened and the resected condylar head was immediately replated into position. A small vitallium plate with 2 small holes at each end was used in the joining of the 2 parts. This plate was fastened to the fragments with vitallium screws. Six months after plating, the monkey was sacrificed. The osteological examination revealed, (a) complete fusion of the fractured head to the neck of the ascending ramus. (b) There are areas of overgrowth of bone which cover part of the vitallium plate. (c) There has been growth in the head of the condyle, forcing the vitallium plate downward onto the ramus. (d) Remodelling has taken place in both the condyle and the neck. (e) There was no evidence of ankylosis, nor was there any indication of gross bony changes on the articular fossa of the temporal bone. (f) The bite did not open, nor was there evidence of malocclusion.

146. A Preliminary Statistical and Morphological Analysis of 425 Cases of Intraoral Squamous Cell Carcinoma. *Richard W. Tiecke, and Joseph L. Bernier, Armed Forces Institute of Pathology, Washington, D. C.* The 405 cases of squamous cell carcinoma of the oral cavity in this study were selected from an original group of 800. Factors which determined their inclusion were completeness of data and adequacy of histologic material. Surgical or biopsy material alone was available in 298 cases, the complete autopsy in 42, and both surgical and autopsy material in 65. Each case was classified

according to location as follows: tongue, floor of the mouth, alveolar mucosa, palate, and buccal mucosa. The mean ages of patients with lesions in these locations were: tongue, 53 years; floor of the mouth, 57 years; alveolar mucosa, 59 years; palate, 57 years; and buccal mucosa, 58 years. Statistical tests indicated that the average age of patients with carcinoma of the tongue was significantly younger than of those with lesions in the other locations. Of 376 patients for whom data on length of survival were available, 44 lived three years or longer. Of these 44 patients 25 (57 per cent) had lesions of the tongue; 3 (7 per cent) in the floor of the mouth; 5 (11 per cent) in the alveolar mucosa; 9 (20 per cent) in the palate; and 2 (5 per cent) on the buccal mucosa. Of the 332 patients who did not survive 3 years or more, 171 (52 per cent) had lesions of the tongue; 57 (17 per cent), floor of the mouth; 39 (12 per cent), alveolar mucosa; 34 (10 per cent), palate; and 31 (9 per cent), buccal mucosa. A survival rate computed by the actuarial method (which takes into account patients who were not followed the full period) for 376 of the patients in the series indicated that 22 per cent could be expected to live 5 or more years. Additional information secured included the duration of the lesion to time of admission, possible etiologic factors, type of treatment, present status of the patient, and metastases wih specific locations. Attempts to correlate degree of malignancy with location, and with other clinical and histological data have not as yet yielded valid information.

XII. Executive Proceedings and Register of Newly Elected Officers and Members

A. COUNCIL SESSIONS

Activities Between General Meetings

Between March, 1951, and March, 1952, the Council determined the time and place of the 1952 general meeting; nominated candidates for the general offices of the Association to be placed on the official ballot-form; accepted the resignation of Dr. S. S. Arnim from the Publication Committee and appointed Dr. Paul E. Boyle to fill the vacancy; created the position of Assistant Secretary-treasurer and selected Dr. Dan Y. Burrill for that position, and approved the promotion of Dr. Marguerite T. Dean to emeritus membership. In addition a correction to the council minutes of the 1951 general meeting was approved, as follows:

On page 525 of the 1951 proceedings (JDR, Aug., 1951), there is the following statement: "A communication was read from Dr. Arthur H. Merritt announcing the incorporation of the William J. Gies Foundation for the Advancement of Dentistry as the successor of the William J. Gies Fund for the *Journal of Dental Research*. Of this fund, Dr. Merritt named himself as president and briefly stated its objectives." The corrected minute reads as follows: A communication was read from the William J. Gies Foundation for the Advancement of Dentistry announcing its incorporation and briefly stating its objectives. The letter was signed by Dr. Arthur H. Merritt, President of the Foundation, and also contains the information that last summer, the William J. Gies Endowment Fund for the Journal of Dental Research became an incorporated body known as the William J. Gies Foundation for the Advancement of Dentistry, Inc.

First Session, Friday, March 21, 1952

In attendance, Doctors Arnold, Boyle, Burrill, Ellis, Fosdick, Hall, Hampp, Hatton, Hill, Hine, Hodge, Kitchen, Kreshover, Manley, Myers, Neumann, Rabinowitch, Speidel and Swanson. Dr. Fosdick called the meeting to order and

called for reports of committees and officers. The Secretary-treasurer submitted the following financial report for the activities of the 1951 calendar year.

Total disbursements	$6,637.77	
Total receipts	5,765.15	
Deficit for 1951	$ 872.62	

Disbursements distributed as follows:

Expenses of office of Secretary-treasurer	$ 629.78	
Expenses of the Publication Committee	173.27	
Reserves for refunds	154.00	
C. V. Mosby Co., JDR account	4,530.72	
Hamilton B. G. Robinson, expense gratuity	750.00	
J. Frank Hall, expense gratuity	250.00	
Edward H. Hatton, expense gratuity	150.00	
Total		$6,637.77

Receipts distributed as follows:

Dues, general fund account	$ 651.65	
Dues, Journal of Dental Research account	3,108.50	
C. V. Mosby Co., JDR expense allowance	1,650.00	
(Balance of $150 included in the 1950 report)		
Gies Foundation for foreign libraries	200.00	
Collections and overpayments	155.00	
Total		$5,765.15

Assets:

General fund,		
Checking account	$1,615.69	
Savings account	1,022.50	
Total		$2,638.19
Endowment Fund,		
Savings bank account	$ 308.26	
U. S. Bonds	1,300.00	
Total		$1,608.26
All assets		$4,246.45

By motion this report was accepted and ordered to be printed in the proceedings. Dr. Hill next reported for the trustees on the task assigned by the Council at the 1951 session (Proceedings, IADR, *J. D. Res.* 30: 525, 1951). This report follows:

The task assigned to me by the Council and Trustees to report on the status of the various endowment funds which have had official or semi-official relationship to this organization, is a difficult one.

The evidence as reported in the Journals of Dental Research is not clear and sometimes appears to complicate what perhaps may be a simple matter. During the existence of the International Association for Dental Research there have been three distinct projects created as endowments in connection with the Association and with the Journal of Dental Research.

In the very first year of the existence of the Journal of Dental Research, gifts were solicited from public spirited persons and institutions, the donors of such gifts were called "fundors" were listed in the Journal of Dental Research,

and the date of the last financial report was 1922. At that time the fund had attained an amount of $6,633.39. On several occasions following that date, notes appeared in the Journal to the effect that efforts to increase the funds had been held in abeyance because of the press of other activities or pending the results of various negotiations. The last of these comments appeared in the Journal of 1932, page iii.

Since the 1932 report there has been no further reference to this fund in the reports of the officers of the Association. The first report of the William J. Gies Endowment Fund for the Journal of Dental Research does not include this item as receipts. It seems probable that this money was expended to make up the constantly recurring deficit in the publication of the Journal.

The second project came into existence in 1933 at the meeting of the IADR in New York on May 25. The Journal for that year records the following statement: "The General Secretary since 1929 presented no bills for expenses in the performance of his duties and made similar arrangements annually for the distribution of reprints of the proceedings to the members during the same period. As these expenses were gifts to the Association, the entire balance in the treasury as of May 25, 1933 ($1,093.70) was converted by unanimous vote into a permanent fund, which it is hoped the Association will be able materially to increase."

The motion called for the transfer of this amount from the general accounts of the Association to be completely segregated as an endowment belonging to the Association and to be used for the support of the Journal of Dental Research, or for other worthy projects. This motion was unanimously approved and the fund authorized. At that time three trustees were appointed to be in charge of this endowment fund and the trustees reported a statement of this endowment fund in the proceedings of the IADR in the Journal, 1935, page 378. Trustees of the endowment fund first appear in 1933. They were B. B. Palmer, Treasurer, for three years; Arthur D. Black, two years, and Russell W. Bunting, one year. No further gifts have ever been made to this fund and this fund with the accumulated interest amounted to $1,608.26 as of Dec. 31, 1951.

The proceedings of the annual meeting of 1933, as of March 14, contains a report of the Trustees in charge of the previous fund (William J. Gies Endowment Fund) but there is no reference to the third project, the Endowment Fund which later that year was announced by Dr. Arthur H. Merritt, called the *William J. Gies Endowment Committee for the Journal of Dental Research.* This article appears in a supplement to the last issue of the Journal for 1936 on page 457. The purpose of this fund as outlined by Dr. Merritt is as follows: "An organized effort is now being made to raise an endowment fund of not less than $50,000 for the Journal of Dental Research. Throughout the seventeen years of its existence, it has served the dental profession with distinction. It was influential above any other agency in obtaining recognition for dentistry as a scientific body by the American Association for the Advancement of Science. More than a thousand research articles have been published on its pages. Its cumulative value to dentistry is beyond computation. And yet, like most research journals, it has never been self-supporting. Since it began publication, in 1919, there has been an average deficit of not less than $2,000. The amounts necessary to meet this recurring debt have been raised by its founder, no small part of which has been contributed by him out of the salary of a college professor.

"There are those in the profession who feel that a situation such as this should not be allowed to continue; that if the profession of dentistry is worthy of the faith which Dr. Gies has shown in it, by devoting more than twenty-five years of his life to its scientific advancement, it will not longer permit him to carry this financial obligation. It is also believed that all that is needed to place

the Journal of Dental Research on a firm financial foundation is to bring the matter squarely before the profession, in order to obtain its wholehearted support. It was with this in mind that the William J. Gies Endowment Committee for the Journal of Dental Research was recently formed. Its purpose is to raise a fund of not less than $50,000, by which it is hoped to place the Journal of Dental Research on a permanent financial basis and, at the same time, make the fund a memorial to Dr. Gies as an expression of appreciation of the service which he has rendered to dentistry."

It is not clear in the Journal who originally appointed this committee or what they originally represented. It says only that Dr. Arthur H. Merritt is the Chairman. In the report at the following annual meeting of this Association on page 360 of the 1937 volume of the JDR, the following statement occurs: "B. *Reports of Officers and Committees.* Council besides reporting routine business indicated at-interim appointment of Drs. Merritt, Chairman, and Eby, Kemple, McKay and Palmer of the New York Section as the *William J. Gies Endowment Committee for the Journal of Dental Research* with power to increase the Committee. . . . *Committee on Endowment.* Committee appointed during the current year and designated the William J. Gies Endowment Committee for the Journal of Dental Research seeks to raise funds to place the Journal on a sound financial basis. Committee's plans include (a) solicitation of contributions from individuals and dental organizations, and (b) endeavor to increase the number of subscribers. Committee proposes to organize cooperative subcommittees in different states. Several organizations already approached and Committee encouraged to believe substantial support forthcoming. Several contributions already received, also new subscriptions—some as 'Sustaining' subscribers at $10.00 per year; some as 'Endowment' subscribers at $25.00 annually; some as 'Life' subscribers at $100.00. Work of Committee going forward satisfactorily. Arthur H. Merritt, Chairman. (Appointment and report approved.)" A later footnote in the Journal of 1938 indicates the following men were additions to that Committee: Theodor Blum, Frank C. Cole, S. Elsworth Davenport, W. B. Dunning, C. Fred GaNum, Gordon M. GaNun, Heman J. Kauffer, Joseph H. Kaufman, Alfred L. Kohn, Richard Pomeroy, Theodor Rosebury, J. A. Salzman, McTracy, Leuman Waugh, and Maurice Williams.

Although the statements as they occur in the published reports in the Journal of Dental Research are not clear on the foundation of this Committee, it appears that the Committee consisted of a group of men who were friends and admirers of Dr. Gies, some of whom were members of the International Association for Dental Research, and some of whom were not. This Committee apparently was organized between meetings of the Association and apparently as an independent entity, although it has had, according to the minutes of the annual meeting of 1937 and at various other times, a formal approval of the IADR, and even the suggestion that it might be a Committee of the Association.

The first report of the Treasurer of the William J. Gies Endowment Fund was made by Dr. Waugh in 1938 as recorded in the Journal, vol. 17, p. 345. The executive proceedings contain this comment: "Report read by Dr. L. M. Waugh in the absence of Dr. Merritt, Chairman. This year's activities along two lines: *First*, planned to create endowment fund of approximately $50,000 by personal letters to key persons. Response gratifying as pledges amounting to about $18,000, more than half of which is paid. *Second*, efforts have been made to substantially increase number of subscribers to Journal. Continued success of Journal depends upon the number of subscribers rather than upon size of any endowment fund. This is highly important and should be and is, one of the main objectives of the Committee having matter in hand. Adopted and referred to the committee of members of New York Section for audit." Thus it would appear that the New York section were the active promoters of the fund.

However, in vol. 18, page 295 of the Journal, in the proceedings of the meeting for 1939 the following statement appears: "The foregoing acts of the Council were approved by subsequent vote of the Association, including election of all nominees for membership listed on pp. 301-303, the adoption of the resolution and approval of the request of the *William J. Gies Endowment Committee of the Journal of Dental Research* to be permitted to invest these funds safely." The report of the Committee for that year showed a fund of $15,275.30. This would appear that the Association had some proprietary interest in this Fund.

Up to the time of the 1951 meeting, the Endowment of the Journal of Dental Research has made annual financial reports to the IADR and these reports have been published as a part of the proceedings of the annual meeting. However, these funds were held in the custody of the Committee and its treasurer. In 1951, this Committee was instrumental in seeing that this fund was incorporated as the *William J. Gies Foundation for the Advancement of Dentistry.* The change in name and organization in incorporation were brought about without asking this Association for its consent and without any form of notification, either by word of mouth or in writing. It does not seem clear at this time that the proprietary interest in this fund was wholly within the IADR or the William Gies Endowment Committee for the Journal of Dental Research. There would appear to be some question as to whether the Committee in this action has kept faith with the donors of this fund whose impression it apparently was (either rightly or wrongly) that this fund would some day become the property of the IADR.

The original prospectus used as a basis for the solicitation of funds very plainly stated that if at any time the Journal of Dental Research became self-supporting, income from the endowment would be used to aid other worthy enterprises included within the general description of the Advancement of Dentistry. It is also very clear that this condition of alternative choices as to the uses for which the funds might be used was a part of the agreement between the donors and the Committee. There is no question but what the Journal of Dental Research is self-supporting if one bases that conclusion on either printed reports of the Publication Committee or personal statements of the members of it. However, it is true that for years after its creation, efforts to make it seem likely that the fund either did or would belong to the Association or its Journal were made.

The impression of the author of this report is as follows: There is no difference of opinion as to the purpose for which this fund was raised, and it can readily be agreed that no matter who administers this fund, either the IADR or the present incorporated Foundation, it will be administered for the best interests of dentistry and for the advancement of its highest ideals. There would seem little doubt that all donors would readily agree that those administering the affairs of this Foundation will use the income of the Fund to the best interests of us all.

Signed, Thomas J. Hill, Chairman.

It was moved, seconded and carried that the report be accepted, the committee thanked for its painstaking exploration of this situation, and discharged. After some minor changes were made it was approved informally, and the report was ordered for publication in the proceedings.

Dr. Hine as chairman of a committee appointed to recommend a time and place for the 1953 meeting reported the results of its investigation. After a number of motions were lost, a motion to hold the meeting in Philadelphia, March 20, 21, 22, 1953 was carried with only one dissenting vote. It was moved and carried that a committee be appointed at an early date to explore the situation for 1955. An informal discussion with reference to the program of the

next general meeting ended without any formal action. Because of the small size and location the sections at Chengtu and Halifax were declared inactive. The Secretary-treasurer was authorized to remove the names of delinquent members from published and mailing lists of members. The dues for 1952-1953 period up to and including Dec. 31, 1953 were set at the rate $7.50 per calendar year for each active North American member. (Motion carried unanimously). Dues of all members living outside of North America were remitted. Disbursements for the 1952-1953 Association year were approved by unanimous vote as follows:

C. V. Mosby Co. for block subscriptions to the Journal of Dental Research at the rate of $6.00 per member subscriber, per year,
For support of the offices of the Secretary-treasurer and Assistant $750.00
For gratuities and expense allowances,

Hamilton B. G. Robinson	$1,000.00
J. Frank Hall	500.00
Edward H. Hatton	300.00
Publication Committee,	300.00

On motion new sections were approved for three key locations in South America (Buenos Aires, Rio Janeiro and Uruguay). Adjourned, to Sunday, March 23.

Second Session, March 23, 1952

Present: Armstrong, Arnold, Boyle, Burrill, Fosdick, Gurley, Hall, Hatton, Hampp, Hill, Hine, Hodge, Kitchen, Kreshover, Manley, Noyes, Rabinowitch, Peyton, Swanson. Approved a list of 92 names submitted by the committee on nominations to membership examined by them and screened for membership qualifications. After adequate discussion these persons were recommended to the Association as fully qualified for membership in the Association. Carried without a dissenting vote. (This list is published elsewhere in these proceedings.) Approved the State Bank and Trust Co. Evanston, Ill., and the Lincoln Bank and Trust Co. of Louisville for deposit of Association funds, withdrawals to be made only on the order of any two of the following officers, President, President-elect and Secretary-treasurer. Authorized the trustees as a committee (Harold C. Hodge, chairman) to select and award a novice oral presentation cash prize of $100.00 for the 1953 general meeting.

Should the participation of guests in the discussions and presentations be limited to a greater extent than has been the practice in the past? This question was discussed at length, it was moved and carried that the Program Committee either adopt some satisfactory plan or if necessary refer the matter to the Council. Dr. Hine moved that if and when The Council on Dental Research of the ADA should invite this Association's participation in its activities by the appointment of a consultant representative this invitation should be accepted. This motion was carried and Dr. Leonard S. Fosdick elected as such consultant, if and when such a position is authorized. (Appointment for one year only). Adjourned.

B. REPORT OF EDITOR—1951

(Volume 30)

During the year, 110 manuscripts were received and of these 63 were published in 1951; 16 have been published in 1952; 16 were returned for revision (the revision of 9 of these being completed) and 15 were rejected.

One hundred original papers were published in Volume 30. In addition, the 896 pages of the volume included 180 abstracts from the Association and

Sectional meetings, proceedings of the Association and of 4 Sections, 6 Editor's Viewpoints and the Index. Promptness of publication is indicated by the "dates received for publication" noted on papers published in the December issue. All of these manuscripts had been received in April, May, June, or August. The quality of papers published appears to be of high order but more frankly unacceptable material is being received, probably reflecting the shortage of space available in other types of dental journals of national circulation.

The pages of the Journal are available for publication of abstracts of papers presented at Sectional meetings. These proceedings should be prepared by the Section Editors, abstracts being limited to 200 words. Papers also presented at the General Meeting of the Association should be recorded by title only in Section Proceedings.

During 1952, it is planned to publish a Directory of I.A.D.R. members in the October issue. This directory, following the style of American Men of Science, has been prepared by Dr. Harold C. Hodge of Rochester.

The Proceedings of the General Meeting are published in this issue.

The statement of the purpose of the Editor's Viewpoint in the April issue, 1951 of the Journal prompted many letters asking for continuation of this feature, and none suggesting its discontinuance.

The Editor wishes to thank the Secretary of the Association, who cooperates in preparation of Proceedings and in many other capacities, the assistants to the Editor, Drs. J. B. Brown and P. C. Kitchin, who generously assist in reviewing manuscripts. The many members who act as anonymous referees, the Ohio State University which supplies his office (and a large part of his time) his secretary, and the contributors and members who make the Journal possible. The Editor also wishes to recognize the invaluable aid and cooperation of the C. V. Mosby Company.

HAMILTON B. G. ROBINSON,
Editor.

C. EXECUTIVE SESSIONS

First Session, Friday, 9:15 A.M., March 21, 1952

The session was called to order by President Fosdick. Members and guests were cordially welcomed and it was announced that because of the heavy snow there would be numerous changes in the printed program. The minutes of the 29th general meeting were approved with one correction and in the form printed in the Journal of Dental Research (30: 455-531, 1951). A brief report was read by the Secretary-treasurer with respect to finances and concerning the activities of the Council between sessions. The balloting for officers was declared in order and Doctors Ned B. Williams, Norman S. Simmons and B. O. A. Thomas were appointed as tellers to receive and poll the ballots. Adjourned to Sunday, March 22, 4:00 P.M.

Session of the North American Divisions, Sunday, March 23, 11:00 A. M.

The meeting was brief as the Association's representative to the American Association for the Advancement of Science holds office until 1956 (Dr. Russell W. Bunting). There was a short report on the last joint session at the holiday meeting of the A. A. A. S. and a preview of the tentative plans for the next meeting with a request for suggestions and advice. Adjourned.

Second and Closing Session, Sunday, March 23, 4:00 P. M.

Meeting called to order by President Fosdick and the tellers reported that 286 approved official ballots were voted and that the list of officers as printed

elsewhere in these proceedings was elected by a nearly unanimous vote. The tellers were thanked and discharged. On motion the Council was authorized to appoint two or more honorary vice-presidents for the coming association year. Dr. Dan Y. Burrill was nominated to the position of Assistant Secretary-treasurer and elected to that position unanimously. The Council proceedings were then brought up for discussion and approval. The recommendation that the 1953 General Meeting be held in Philadelphia in March received a generous discussion both for and against the proposal, but was carried on the motion to that effect by a hand vote. Dues for the 1952-1953 period up to and including Dec. 31, 1953 were set at the rate of $7.50 per calendar year for North American members, and remitted for members residing outside of North America. Disbursements for the period were authorized as listed in the Council proceedings. The list of persons nominated for membership as approved by the Council and as having met all qualifications for membership was read. On the basis of unanimous approval, the rules of procedure were suspended and a written ballot formally cast for the 92 candidates on this list and as printed elsewhere in these proceedings. The president then declared them elected to membership. The Lincoln Bank and Trust Co. of Louisville and the State Bank and Trust Co. in Evanston were authorized by the regular procedure to handle Association funds and to make disbursements on the written order of any two of the three following officers: President M. K. Hine, Secretary-treasurer Edward H. Hatton, and President-elect Francis A. Arnold, Jr. All other activities of the Council were approved and the Council was given authority to act for the Association during the interim between this and the next general meeting. Mr. Richard Scott was warmly thanked for his excellent services in connection with slide projection. Also the local committee, Dr. Orban, chairman and Mrs. Fosdick and Mrs. J. R. Jarabak for their help with the registrations. A personal message from Dr. Wm. J. Gies was read and the Secretary-treasurer directed to remind him again of the Association's great gratitude for his unfailing interest in the Association and his long service to it.

Since the last general meeting, the Association has lost by death Peter J. Brekhus, James E. Cary, M. Don Clawson, William L. Fickes, Philip A. Leavitt, Moe K. Marcus, E. B. Owen, Walter H. Scherer, James T. Sweeney, Benjamin Tischler, G. R. Winter, Walter H. Wright, James L. Zemsky. At the request of President Fosdick, members and guests stood briefly and in silence as a tribute to these former members.

The newly elected officers were then called to the front by President Fosdick and inducted informally into their respective offices. Incoming President Hine then accepted the gavel from the outgoing president and addressed the Association briefly, his remarks received close attention and at the close he was warmly applauded. There being no further business the 1952 general session was then adjourned at 5:30 P. M.

D. OFFICERS AND MEMBERS ELECTED

ACTIVE OFFICERS: President—*Maynard K. Hine*, University of Indiana. President-elect—*Francis A. Arnold, Jr.*, National Institute of Dental Research. Vice-president—*George C. Paffenbarger*, National Bureau of Standards. Secretary-treasurer—*Edward H. Hatton*, Northwestern University. Assistant Secretary-treasurer—*Dan Y. Burrill*, University of Louisville. Trustee for a five-year term—*Thomas J. Hill*, Western Reserve University. Editor—*Hamilton B. G. Robinson*, Ohio State U. Members of the Publication Committee—*J. Frank Hall*, University of Indiana and *Paul E. Boyle*, University of Pennsylvania.

MEMBERS ELECTED. FROM NORTH AMERICA: Alabama—*L. Rush Bailey, Clarence E. Klapper, Ward Pigman and Arthur H. Wuehrmann*, University of Alabama. California—*Earl Gilmore, Ernest F. Inskipp, E. L. Johnson, Paul R. Thomassen* and *John K. Young*,

College of Physicians and Surgeons. Connecticut—*Harold S. Fleming*, Yale University; *Elizabeth A. Luster*, J. M. Ney Co., District of Columbia—*Theodore E. Fisher*, National Bureau of Standards; *Irving B. Fritz*, Army Med. Service Graduate School; *Joseph L. Henry*, Howard University; *Gustav O. Kruger*, Georgetown University; *Ralph Lobene*, Army Dental Research Laboratory; *Herman Medak*, Army Medical Service Graduate School; *Denton L. Smith*, National Bureau of Standards; Illinois—*Benjamin J. Gans*, Loyola University; *Kermit F. Knudtzon; Eli Olech*, University of Illinois; *Karl A. Piez*, Northwestern University; Iowa—*Paul Belding*. Kentucky—*William R. Wolfe*, University of Louisville. Massachusetts—*Paul W. Munson*, Harvard University; *Samuel S. Turesky*, Tufts College; *Naomi C. Turner*, Forsyth Dental Infirmary. Maryland—*Charles W. Devier* and *Hubert B. Palmer*, University of Maryland. Michigan—*Spencer H. Bush* and *Sigurd Ramfjord*, University of Michigan. Minnesota—*James R. Jensen*, and *William A. Peterson*, University of Minnesota. New Jersey—*Arnold Singer*, Block Drug Co. New York—*Dimitri A. Afonsky*, University of Rochester; *Saul M. Bien; Solon A. Ellison*, and *Paul Goldhaber*, Columbia University; *Robert C. Hockett*, Sugar Research Foundation; *Sidney L. Horwitz*, Columbia University; *Edward Klein, S. Sigmund Stahl* and *Otto Trautz*, New York University. Ohio—*Robert E. Gillis* and *Grace C. Kimball*, Western Reserve U. Oklahoma—*Wayne T. White*, Ozark-Mahoning Co. Ontario (Canada)—*H. A. Hunter, Gordon Nikiforuk* and *James J. Rae*, University of Toronto. Oregon—*Bhim Sen Savara* and *Theodore Suher*, University of Oregon. Pennsylvania—*S. Wah Leung, Jessica W. Nathans* and *Lester H. Roth*, University of Pittsburgh; *Charles E. Wilde, Jr.*, University of Pennsylvania. Tennessee—*Charles C. Chumbley; Faustin N. Weber*, University of Tennessee.

MEMBERS ELECTED FROM COUNTRIES OUTSIDE OF NORTH AMERICA. Australia (Sidney)—*Neil E. Goldsworthy*, Institute of Dental Research. Argentina (Buenos Aires)—*Jorge E. T. French* and *Rosa Iusem*. Brazil (Rio de Janeiro)—*Sylvio Bevilacqua, Mario de Magalhaes Chaves* and *Claudio Mello*. Denmark (Copenhagen)—*Arne Björk, Knud D. Jorgensen, Gertrud Lund* and *Marie Ussing*, Royal Dental College. England (Birmingham)—*J. L. Hardwick*, Dental Hospital; *E. B. Manley*, Medical School. (Bristol)—*A. R. Darling*, Dental Hospital. (Cambridge)—*Mrs. S. Hughes*. Harwell, Atomic Energy Research Establishment. (Liverpool)—*R. L. Hartles, A. S. Prophet, G. L. Slack, H. H. Stones* and *R. D. Emslie*, School of Dental Surgery. (London)—*I. R. H. Kramer*, Eastman Dental Hospital; *A. B. MacGregor; A. E. W. Miles*, London Hospital Dental School; *G. F. Parfitt*, Eastman Dental Hospital; *M. V. Stack*, Guy's Hospital. (Manchester)—*H. F. Atkinson* and *E. Mathews*, Turner Dental School; and *J. Miller*, Dental Hospital of Manchester; (Newcastle upon Tyne)—*R. V. Bradlaw*, Sutherland Dental School; *G. N. Jenkins* and *D. E. Wright*, Medical School, King's College. (Sheffield)—*E. W. Bradford*, and *J. J. Hodson*, School of Dental Surgery, The University. Sweden (Stockholm)—*Allan Stralfors*. Switzerland (Basle)—*Roland Bay*.

RESIGNATIONS DURING THE 1951-1952 ASSOCIATION YEAR. K. C. Christensen, H. T. Fraser, Brooks Juett, Paul Lalonde, Samuel J. Lewis, Duane W. Lovett, Edith Oblatt, L. W. Peterson, E. O. Shaner and Eugene R. Stone.

E. MEMBERS VOTING AT THE ANNUAL ELECTION*

Aisenberg, Allen, G. M. Anderson, Applebaun, Archer, Armstrong, Arnold, Ash, Bagnall, C. R. Baker, Baume, Bender, Berger, Bernier, Bhaskar, Blackberg, Blackerby, R. E. Blackwell, Blayney, Jack Bloom, Blumenthal, Bodecker, Boucher, Boulger, Boyle, Brand, Brandhorst, Brashear, Broadbent, Brodie, Brodsky, Brucker, Bryan, Burkett, Burnett, Burrill, Burstone, Canby, Carr, Caul, Chase, Chernausek, Cheyne, Chilton, Clark, Clausen, Clough, M. M. Cohen, T. B. Coolidge, Costich, Cox, Coy, Cross, Crowley, Dahlberg, Dale, Darlington, Davis, H. T. Dean, Dickson, Dietz, Dobbs, Dragiff, Dreizen, Dressell, Dummett, J. M. Dunning, Easlik, East, Easton, F. G. Elliott, Ellis, Elvove, English, Ennever, Ennis, Sidney Epstein, Erikson, Everett, Ferguson, Finn, A. K. Fisher, Fleming, Forbes, Fosdick, Freeman, Frey, Frisbie, Gafafer, Gamboa, Gilchrist, Gilda, Gilkison, Ginn, Glass, H. J. V. Goldberg, Goldman, Greep,

*Because of failure of a few members to identify envelopes enclosing ballots, the names of some voting members cannot be supplied.

Grosby, Grossman, Gruebbel, Gurley, Hadley, Hahn, Hall, Harman, Hatton, Hein, Hess, I. N. Hill, T. J. Hill, Hine, Hodge, Hoffman, Hooper, Hubbell, Hunt, Ireland, Jacobson, James, Jarabak, Jasper, Jeserich, L. G. Jordan, Kaiser, Kaletsky, Kaplan, Karshan, Kerr, Kesel, D. A. Keys, Kitchen, Klaffenbach, Klatsky, Kniesner, Knighton, Koser, Kreshover, Krogh, Kurth, Lalonde, Land, Lasater, Law, Lefkowitz, Levy, Lieban, Losee, Lovestadt, P. G. Lowery, Ludwick, Lundquist, MacDonald, Mahler, Manley, Mann, Margolis, Marjerison, Marshall-Day, Massler, Mead, Merkeley, Mercado, Merritt, Midgeley, Milhon, S. G. Miller, Miner, Mitchell, Moen, N. A. Moore, T. E. Moore, Anna Morse, F. W. Morse, Jr., A. H. Mueller, Emil Mueller, T. F. McBride, McCauley, McCollum, McCrea, McKay, Neumann, Neuwirth, R. J. Nelsen, Clyde Nelson, Nuckolls, Oliver, Orban, Orland, Ostrom, Paffenbarger, Parke, Patterson, Pearlman, Permar, Peterson, Peyton, Pfeiffer, Phillips, Podolin, Prinz, Rabkin, Randall, Rankow, Rault, Reveszz, H. B. G. Robinson, Romnes, Rosenstein, Rovelstad, Rudolph, Russell, Ruzicka, Ryder, Sarnat, Savchuck, Schantz, Schlack, Schoonover, Schour, D. B. Scott, Scrivener, Shafer, Shapiro, Shaw, Shell, Sherman, Sicher, Siegel, Simmonds, Simon, Sloman, E. L. Smith, Smyd, Snyder, Sobel, Sognnaes, Souder, Spangenberg, Speidel, Stafne, Georg Stein, Steinmeyer, T. D. Stewart, Strader, H. A. Swanson, W. F. Swanson, Swartz, Taber, N. O. Taylor, W. C. Taylor, Thoma, B. O. A. Thomas, H. S. Thompson, J. R. Thompson, Tiecke, Tilden, Van Huysen, Van Kirk, Vehe, Volker, Volland, Wach, Waggener, Wainwright, Wallace, Walter, Ward, Waugh, Weinmann, Wentz, Wessinger, Wheatcroft, Wheeler, Wilbur, Williams, Willman, Wolcott, F. J. Wolfe, Wright, Wylie (Cal.), Zipkin. Total, 283.

F. MEMBERS IN ATTENDANCE AT ONE OR MORE SESSIONS, LISTED BY SECTIONS

Ann Arbor—Bunting, Jeserich, Peyton, Vedder. *Baltimore*—McCrea. *Birmingham*—Finn, Volker. *Boston*—Barr, Glass, Greep, Keyes, Kraus, Lisanti, Manly, Marjerison, Marshall-Day, Shiere, Sognnaes, Yurkstas. *Chicago*—Blayney, Brodie, Burstone, E. D. Coolidge, T. B. Coolidge, Engel, Fosdick, Freeman, Harrison, Hatton, Hill, Jarabak, Koser, Orban, Orland, Pearlman, Rapp, Rovelstad, Schour, Sicher, Skinner, Tylman, Wainwright, Wallace, Weinmann. *Cleveland*—Hill, Scheu, Wylie. *Columbus*—Kitchin, Permar, Spangenberg. *Indianapolis*—Boyd, Hall, Hine, McDonald, Phillips, Shafer, Van Huysen. *Iowa City*—Boyd, Smith, Wessels. *Kansas City*—Moore, Myers. *Louisville*—Burrill. *Minnesota*—Armstrong, Clark, Crawford, Lovestedt, Mitchell, Savchuck, Speidel, Zander. *Nebraska*—Hooper, Ireland. *New York*—Benzer, Neumann. *Pacific-Northwest*—Jump, Law, Noyes, Phatak, Snyder, Thomas. *Philadelphia*—Boyle, Burkett, Crowell, Taylor, Williams. *Pittsburgh*—Cox, Swanson, Van Kirk. *Richmond*—Coy, Kreshover, Lyons. *Rochester*—Bibby, Brudevold, Costich, Hein, Hodge. *San Francisco*—Agnew, Baume, Becks, Epstein, Frisbie, Nuckolls, Ray, Scrivener, Wylie. *St. Louis*—Allen, Boling, Brandhorst, Dietz, Gurley. *Tennessee*—Ginn. *Toronto*—Ellis. MacDonald. *Washington*—Arnold, Burnett, Caul, Dean, Dickson, East, English, Glasson, Hampp, McBride, Nelsen, Oartel, Paffenbarger, Parke, Rault, Rogosa, Schoonover, Scott, Stephan, Sweeney, Tiecke, Wheatcroft, Zipkin.

Not Belonging to a Section—Brauer, Breitner, Deakins, Hunt, Molnar, T. E. Moore, Nelson, Pindborg, Ritchey, Ruge, Smyd. Total, 148.

Attendance of New Members, Elected March 23, 1952—Bailey, Bush, Fischer, Henry, Hockett, Inskipp, Knudtzon, Leung, Klapper, Kruger, Munson, Piez, Pigman, Rae, Ramfjord, Savara, Stones, Stralfors, Suher, Wuehrmann. Total, 21.

Total registered attendance of members and guests, 359.

THE EFFECT OF THE CHANGING DENTITION ON MASTICATORY FUNCTION

F. R. SHIERE AND R. S. MANLY

From the Oral Pediatrics Department and the Laboratory for Oral Physiology, Tufts College Dental School, Boston, Mass.

INVESTIGATIONS on the development of masticatory function among children have usually been concerned with the eruption times for teeth and the maximum biting force exerted on molars or groups of teeth. Dennis[1] reported a range of 10 to 20 pounds in maximum biting force for children 8 to 14 years of age. Johnson and Hatfield[2] studied forces on changing dentitions and observed the greatest increase in biting strength from 11 to 13 years of age with abnormal dentitions showing lower average forces. Lancet[3] reported an average maximum biting force of 57 pounds at 7 years of age, which decreased with age up to 10 years, increased to 12 years, and decreased again as the second molar erupted. Friel[4] studied incisor forces and made comparisons with forces exerted by the hand grip. Brawley and Sedwick[5] gave data on 783 school children from 6 to 17 years of age, which showed a gradual increase of 5 pounds per year, reaching the mean value of 83 pounds at 17 years of age. Studies by Worner and Anderson[6] included 300 children from 4 to 18 years of age, in which they reported that the biting force on the first permanent molar increases at the rate of 3 pounds per year over the age range of 7 to 16 years.

No information on the change of chewing ability with age has been reported as far as the authors are aware. This study was planned to establish normal ranges of masticatory ability among children from 6 to 15 years of age and to evaluate how the changing dentition and the loss of teeth influences masticatory ability. A practical question which might be answered is whether or not teeth lost through dental caries should be replaced by space maintainers that have a functional occlusal surface.

EXPERIMENTAL PROCEDURE

Two hundred children, 6 to 10 years of age, were obtained for study at the Department of Oral Pediatrics, Tufts College Dental School, and the same number, age 6 to 15 years, were examined at the Pond Plain School at Westwood, Massachusetts. Each child was given several tests for masticatory ability and readings of maximum biting force were taken on all molar teeth, using the instrument described by Howell and Manly.[7] Each subject was tested for food platform area[8] in duplicate on each side of the mouth.

During the preliminary phases of the work, mastication tests were performed with peanuts and carrots as test foods on each side of the mouth. Each food was tested first with 20 masticatory strokes per 3 Gm. portion, and secondly

This investigation was supported in part by a research grant from the National Institute for Dental Research of the National Institutes of Health, Public Health Service.

Read at the 30th General Meeting of The International Association for Dental Research, Colorado Springs, March 23, 1952.

Received for publication, Feb. 10, 1952.

with whatever number of strokes the child judged he needed in order to prepare the food for swallowing. This latter procedure has been termed the "swallowing threshold."[9] After the first hundred cases had been studied, it was discovered that the masticatory performance results with the two foods were very well correlated, and that it would therefore be unnecessary to continue the tests with carrots. The same held true for the tests on swallowing thresholds. Tests with carrots were eliminated and replaced by an additional peanut test, using either 60 or 80 masticatory strokes; this additional information permitted a calculation of masticatory efficiency.

RESULTS

Correlations Among Function Tests.—In previous studies with natural dentition cases[10] and full denture wearers,[11] it was found that the masticatory performance with the soft food (peanuts) was well related to the chewing ability on the tougher food (carrots). The two parameters seem to measure the same phase in mastication and results with both foods are not needed. For this reason, the findings from the first 100 cases were examined in order to learn whether the same close correlation would be found for children. The correlation coefficient between the masticatory performance with peanuts (P_p-10) and with carrots (P_c-5) was found to be 0.86. A similar high correlation was found between the swallowing threshold with peanuts (U_p) and with carrots (U_c). Here the coefficient was 0.95. In view of these high correlation coefficients it was considered unnecessary to continue with carrot tests, since the results with softer foods would serve as indices of chewing ability.

In a previous publication[10] methods for calculating masticatory efficiency were based upon the number of chews required by a subject in order for him to be able to achieve a standard degree of pulverization. Sometimes this number could be obtained by interpolation. For example, if a subject were found to be capable of pulverizing 76 per cent of the peanuts to pass through a 10 mesh screen at 28 chews, and 80 per cent at 32 chews, 30 strokes would be required for the "standard" degree of pulverization, 78 per cent. When the strokes required are divided into 20 (the strokes required by the standard person), the efficiency of mastication for the unknown person is found to be 67 per cent. The interpolations were actually performed on a chart which had pulverization on a probability scale, and the number of chews on a logarithmic scale. A similar type of efficiency calculation could be obtained from data at 20 and 60 chews. In this instance, however, most of the children were found to have efficiencies lower than 100 per cent, and it was desirable to use a different base line in making the calculation. This base line was 65 per cent through a 10 mesh screen which corresponded to 62 per cent efficiency. From these interpolations the "actual" efficiencies were determined for 170 dentitions.

The efficiency could also be calculated by prediction from the performance at 20 masticatory strokes. This method has already been published.[12] The correlation between predicted and actual efficiency is presented in Table I. The

TABLE I

CORRELATION BETWEEN "ACTUAL" EFFICIENCY AS DETERMINED FROM TESTS AT 20 AND 60 MASTICATORY STROKES, AND "PREDICTED" EFFICIENCY THAT IS CALCULATED FROM TESTS AT 20 CHEWS BY TABLE II OF REFERENCE 10. THE CONTINGENCY COEFFICIENT IS HIGH (0.907)

		PREDICTED EFFICIENCY									
		1-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100
Actual efficiency	1-10	2	1								
	11-20	3	11	6		1					
	21-30	2	12	15	4	1					
	31-40		1	6	17	10					
	41-50			4	15	11	5				
	51-60				2	2	10				
	61-70							15			
	71-80							2	3	2	
	81-90									5	
	91-100							1			

correlation coefficient is .907 which is high enough to indicate that for children the masticatory efficiency can be predicted with considerable precision from the masticatory performance test.

In previous investigations dealing with natural dentition cases,[10] denture wearers,[11] and orthodontic cases,[13] it was found that the particle size of the food at swallowing is directly related to the masticatory performance. The data on 279 half dentitions have been compared in Table II, which demonstrates the relationship between masticatory performance at 20 strokes (P_p-10) and the swallowing threshold (U_p). Here the relationship is linear and is well correlated; the correlation coefficient has been found to be .796. This finding indicates that a defective masticatory ability causes the child to swallow larger particles of food than he would if his ability were normal. These high correlations indicate that the masticatory efficiency is a suitable single index of masticatory function, since it is related in a complicated mathematical fashion to the masticatory performance, which in turn is directly related in a linear fashion to the swallowing threshold.

TABLE II

RELATIONSHIP BETWEEN P_p-10 AND U_p. THE PARTICLE SIZE OF FOOD AT THE TIME OF SWALLOWING IS WELL CORRELATED WITH THE CHEWING ABILITY OF THE CHILD (CONTINGENCY COEFFICIENT = 0.796)

		SWALLOWING THRESHOLD									
		1-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100
Performance	1-10	1	1								
	11-20		4	3		1	1				
	21-30	1	5	6	10	6	1				
	31-40				9	7	12	7	2	1	
	41-50				6	11	11	11	8		
	51-60				1	3	14	26	12	2	1
	61-70				1	2	9	18	14	8	1
	71-80					1	1	2	14	6	2
	81-90					1			4	5	2
	91-100								1	1	

This finding permits a simplification of treatment of the remainder of the data. There were four variables included in the study which were independent in the sense that any one might influence masticatory function: these were age, sex, numbers and types of teeth in occlusion, and degree of malocclusion. There were five separate measures of function: masticatory efficiency, masticatory performance, swallowing threshold, maximum biting force, and the food platform area. Food platform area was not highly correlated with masticatory efficiency. Since masticatory performance with peanuts (P_p), and carrots (P_c) and the swallowing threshold (U_p) were closely related to efficiency, it was not necessary to make separate comparisons between the first two measures of function, and the four independent variables.

Influences of Independent Variables Upon Masticatory Function.—Loss of primary and permanent teeth appears to have some effect on the masticatory ability of children, but the range in this effect is quite large. Table III shows the distribution of masticatory efficiencies for half dentitions with all teeth present, and for those lacking the first primary molar, the second primary molar, or the first permanent molar. An extremely wide range of efficiency is possessed by children who have all teeth present (11 to 100). The median efficiency for the group with no missing teeth occurs in the 41 to 50 bracket, or about half of that possessed by a "standard" dentition, which also has a second permanent molar in occlusion. With the first primary molar missing the range is about the same as before, but the median occurs in the 30 per cent group.

TABLE III

DISTRIBUTION OF MASTICATORY EFFICIENCIES FOR THE PREFERRED SIDE OF THE DENTITION

	EFFICIENCY									
	1-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100
All teeth present	1	15	19	29	38	21	16	12	7	5
One first permanent molar missing			4	1						
One second primary molar missing			7	2	1					
One first primary molar missing		6	5	5	6		1	1	1	

For examination of the influence of age on masticatory function, cases were selected which had all teeth present. Only data from the preferred side were used, since this side would be most practiced in food manipulation. The results are presented in Fig. 1. Average masticatory efficiency for each age is represented by the height of the columns and the age is indicated along the abscissa. The vertical line at the top of each column shows the standard error of the mean, and the number within the column refers to the number of cases. Children six years of age are divided into groups, with and without eruption of the first permanent molar.

It is clear that eruption of the first permanent molar does not immediately improve mastication; there is no statistical difference between the efficiencies of six-year-old children with the first permanent molar in occlusion and those with unerupted first molars. The differences in efficiencies of 7, 8, and 9-year-

old children are approaching significance (90 per cent level), and efficiencies for children 9 years of age are definitely higher than those at 7 years (99 per cent). Because of the wide scatter of data, the 10 year old cannot be proved significantly different from 9-year-old children, but is definitely superior to those 8 years of age (99 per cent level). Thus each succeeding year from age 7 to age 10 appears to bring about gradual improvment in chewing ability.

At age 11 the efficiency falls (95 per cent level of significance) and remains about the same through age 13. Once again it starts to rise, since the 14 year old averages higher than the 13-year-old children (99 per cent level). Few cases are reported for ages 11 and 12, because most of the half dentitions studied had at least one tooth missing due to exfoliation and could not be included in Fig. 1. The lower efficiencies through ages 11 to 13 probably are attributable to the time required for newly erupted teeth to become functional. The substitution of premolars for primary molars has the immediate effect of reducing efficiency by one third. The eruption of a second molar at age 12 or 13 does not raise efficiency immediately. From one to three years after these teeth have been in function, the efficiency rises to approach the level which had been reached with the original primary dentition.

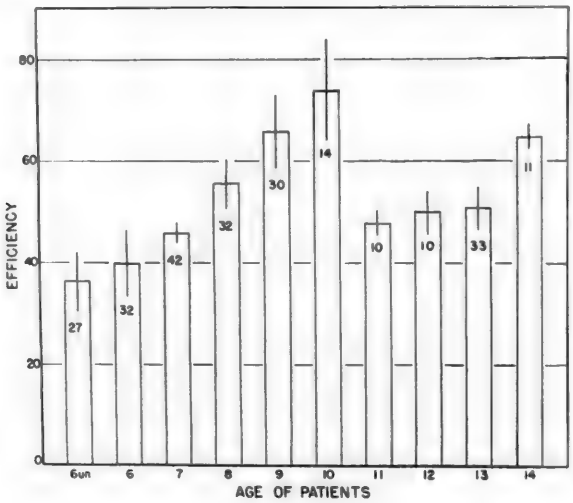


Fig. 1.

The efficiency of adults with dentitions that are complete except for third molars have been used as the base for 100 per cent efficiency.[12] Children 12 or 13 years of age with the same teeth in occlusion have an average efficiency that is only half as great. At age 14 it becomes two-thirds as great. Probably 100 per cent efficiency would be reached at age 16 or later. This means that teeth must be in function for three years or longer before their maximum utility in mastication is reached. This conclusion checks with the observations on the first permanent molar.

During the course of the study a total of 187 cases were observed which would be classed as malocclusion. It was noted that the seven Class III cases had very low areas and performances, and the two measures of function ran hand in hand. In order to compare the influence of malocclusion on chewing ability, a graph was prepared (not shown) in which masticatory performance was plotted against the food platform area with provision for separate observation of Class I, Class II, Class III, and crossbite cases. It was found that there was no tendency for the information to cluster on this chart. Hence all classes of malocclusion were similar in their ability to use the food platform area which was available to them.

The question of whether males and females differed in masticatory ability was studied by comparison of groups of boys and girls who were similar in age, who had a limited range in area (8 to 27 mm.^{2}), and who possessed the same numbers of teeth in occlusion. Two comparisons were made. In one comparison the sexes had ages of 12 to 14 inclusive and had two permanent molars and two premolars in occlusion. The mean performance for males was 44 and for females was 41. This difference was not statistically significant. In the other comparison, the ages were 7 to 9 inclusive with one permanent first molar and two primary molars present. The mean performance for boys was 51 and for girls it was 47. Here again the difference was not significant, even though the males again showed a higher average figure.

It has been shown by Johnson and Hatfield[2] that the maximum biting force of individuals steadily increases with age. It seems important to examine whether the same relationship would hold with our data using average forces on molar and premolar teeth. At the age of 6 the average force for 24 children with unerupted first permanent molars was 44.7 ± 1.7 (s.e.) pounds, whereas that for the 10 half dentitions with the first molar erupted was 51 pounds. At age 13 the average force was 52.5 ± 0.8. The ranges for the intervening ages were wide and means differed no more than would be expected from chance. The changes from age 6 to 13 were not as great as would be expected from the previous literature on this subject.

Other Factors Influencing Masticatory Function.—In a previous publication[12] the food platform area was found to be well correlated with the masticatory performance and masticatory efficiency among young adults. In this study, area could be compared with masticatory performance for 800 half dentitions obtained from tests on children. The results are presented in Table IV. There is a correlation with masticatory performance, but the coefficient is not as high as was found in previous studies. The coefficient of mean square contingency for these groups of data was found to be .567. With a coefficient of this magnitude there is little predictive value for masticatory efficiency from the area. Part of the reason for the lower coefficient may be attributable to the diminished range in the values; the mean area for adults with a natural half dentition lacking only a third molar was found to be 35 sq. mm., whereas 90 per cent of the children have less area.

The data which does most to detract from the magnitude of the correlation is found in the upper right quadrant of Table IV; it comprises those cases with

efficiencies of 28 or over, and areas of less than 7 sq. mm. Perhaps certain groups of cases contribute more than others to this group of deviating data. In order to test this hypothesis, the performance was plotted against the area for different ages of children keeping the complete dentitions separate from those with missing teeth and the preferred side separate from the nonpreferred side. The distribution of points were similar when the preferred and nonpreferred sides were compared and when cases with all teeth present were contrasted with

TABLE IV

RELATIONSHIP BETWEEN FOOD PLATFORM AREA AND MASTICATORY EFFICIENCY. THE CORRELATION BETWEEN THESE VARIABLES IS ONLY MODERATE (0.567)

		PREDICTED EFFICIENCY									
		1-10	11-20	21-30	31-40	41-50	51-60	61-70	71-80	81-90	91-100
Area	0-3	2		9	3	2	2				
	4-7	2	11	8	5	6	1		1	1	1
	8-11	2	14	17	16	15	4		4	2	3
	12-15		25	34	28	18	7	6			5
	16-19		7	17	9	21	8	6	3	3	4
	20-23		2	4	12	14	11	7	10	2	1
	24-27		1		6	10	5	6	2	3	3
	28-30				4	4	3	4	3	3	5
	32-35					4	1	2	1		2
	36-39							3		2	1

those having some teeth missing. However, at ages of 8 to 10, 15 per cent of the cases fell in high performance-low area range as compared with 5 per cent or less at all other ages. This indicates that area is apt to be of less predictive value at ages 8 to 10 than at other ages, and that low area does not necessarily mean low performance at these ages.

As in previous studies on adults, the maximum force was found to have little relationship to performance. The averages of maximum biting forces on molar and premolar teeth was from 30 to 60 pounds. There was no evidence of a correlation with masticatory efficiency. The relationship between maximum force and food platform area was found to result in a triangular scatter of points indicating that persons with high forces might either have a high or low area but that cases with low force did not have high area and cases with a high area did not have a low force. There are several possible interpretations of this fact, but none deserve discussion since the experimental fact is of little usefulness in explaining why one child differs from another in ability to masticate food.

In a previous study with 50 adult patients,[14] it was found that persons will usually prefer the better of the two sides of the dentition for chewing when the difference is great, but will usually select the right side when the two half dentitions are about equal in masticatory performance (±10 per cent). In this study the range of equal performance was considered to be ±20 per cent. There were 315 cases for whom the two sides of the mouth differed by 20 per cent or less. Among these, 205 cases preferred the right side in mastication, 96 preferred the left, and 14 had no preference. Here again is an indication that the right

side is generally preferred when masticatory ability is approximately equivalent. There were 28 cases where the right side was 20 per cent better than the left side. Among these, 23 of the cases preferred the right side and four had no preference. There were 27 cases with the left side 20 per cent greater than the right side in performance; among these, 13 preferred the left and 13 preferred the right and one had no preference. Among children the tendency to prefer the right side predominates over the tendency to prefer the more efficient side.

SUMMARY

An investigation has been made of 400 children from 6 to 15 years of age in order to determine the effect of the changing dentition on masticatory function. Masticatory performance, swallowing threshold, food platform area and the maximum biting force were determined on both sides of the dentitions. It was possible to predict masticatory efficiency from masticatory performance with considerable accuracy. The masticatory performances with peanuts, P_p, were well correlated with the performances with carrots, P_c, with efficiency, E_p, and with the swallowing threshold, U_p.

The masticatory efficiencies of the children extended over a wide range and averaged about half that of adults who had all teeth except the third molar present. The first and second permanent molars did not improve masticatory function immediately after eruption, but efficiency gradually increased over a period of three to four years. Efficiency declined after age 10 and rose again by age 14, probably because of the time required for the recently erupted teeth to become functional.

Forty-seven per cent of 400 cases investigated were classed as malocclusions by orthodontic criteria. Only the Class III cases showed a low area and a corresponding low performance. Other malocclusions were similar to normal cases in their average masticatory function. There was no significant difference in the efficiency of males and females with similar ages and a limited range in area. Food platform area was less well correlated with efficiency (.57) than was found previously among adults.

The maximum biting force had little relationship to masticatory efficiency or food platform area. The tendency to prefer the right side in mastication predominated over the tendency to prefer the more efficient side in mastication.

The authors wish to acknowledge the cooperation of Mr. Edmund Thurston, Superintendent of Westwood Schools, in arranging for observations to be made on two hundred Westwood children. Technical assistance was provided by Mrs. Helmut Zander and Miss Verna Crowell, and statistical work was performed by Mrs. Joan Keyser.

REFERENCES

1. Dennis, George J.: A Study of the Masticating Force of the Jaws, *D. Cosmos* **35**: 1045, 1893.
2. Johnson, A. L., and Hatfield, H. K.: A Study of the Relation of Dental Conditions, Biting Force, and the Hand Grip, *D. Cosmos* **59**: 599, 1917.
3. Lancet, B.: An Investigation into the Relationship of the Facial Musculature to the Hand Pressure and Vital Capacity, *D. Cosmos* **69**: 569, 1927.
4. Friel, E. S.: Muscle Testing and Muscle Training, *D. Record* **44**: 187, 1924.
5. Brawley, R. E., and Sedwick, H. J.: Studies Concerning the Oral Cavity and Saliva. Measurements of Biting Pressure in Children, *A. J. Orthodont.* **26**: 41, 1940.

6. Worner, H. K., and Anderson, M. N.: Biting Force Measurements On Children, *Australian J. Dent.* **48**: 1, 1944.
7. Howell, A. H., and Manly, R. S.: An Electronic Strain Gauge for Measuring Oral Forces, *J. D. Res.* **27**: 705, 1948.
8. Yurkstas, A., and Manly, R. S.: Measurement of Occlusal Contact Area Effective in Mastication, *Am. J. Orthodont.* **35**: 185, 1949.
9. Yurkstas, A.: Compensation for Inadequate Mastication, *Brit. D. J.* **91**: 261, 1951.
10. Manly, R. S.: Factors Affecting Masticatory Performance and Efficiency Among Young Adults, *J. D. Res.* **30**: 874, 1951.
11. Manly, R. S., and Vinton, P.: Factors Influencing Denture Function, J. Pros. Den. **1**: 578, 1951.
12. Manly, R. S., and Braley, L. C.: Masticatory Performance and Efficiency, J. D. Res. **29**: 448, 1950.
13. Manly, R. S., and Hoffmeistr, F. S.: Masticatory Function of Orthodontic Children, *J. D. Res.* **30**: 474, 1951.
14. Manly, R. S., and Shiere, F. R.: The Effect of Dental Deficiency on Mastication and Food Preference, *Oral Surg., Oral Med., and Oral Path.* **3**: 674, 1950.

VARIATIONS IN STRUCTURAL MORPHOGENESIS OF THE HUMAN FACE AND JAWS

Initial Stages of Normal and Abnormal Bone Formation (Gross) Exhibited in the Cleared Embyro and Fetus

SAMUEL RABKIN

Winter Park, Florida

IT IS common knowledge that variations in the physical morphology of man's face and jaws have prevailed throughout his known history. Random mating and territorial changes no doubt account for the like and unlike characteristics of the head and face of ancestral man. Deviations from normal, either in anatomic configuration or opposing jaw relationships, infrequently occurred among prehistoric man and more recent primitives. However, the familiar deformities, distortions, and nonuniformities that exist at present followed in the wake of advanced civilization (Fig. 1).

Markedly contrasting bone formation is also present in modern man. A wide range of variations and extremes, from the thick, massive, and dense compact walls with densely deposited spongiosa, contrasts with poorer quality, light weight, thin, frail, and porous bone. These conditions are found when skeletal material is examined. X-rays and enlargements made of gross sections cut transversely and longitudinally disclose greater detail in the trabecular structure between markedly dense layers and the sparsely "hypocalcemic" ossification. Though these conditions prevail in the long bones and the walls of most of the cranial vault, it is the thin and delicately formed bone comprising the maxilla, palate, and adjacent sinus walls that are invariably predisposed to local degenerative changes and disease. In such instances the alveolar process suffers most.

X-rays of crania of the frail and porous type, cut sagitally through the midline, often disclose no more than a hazy area of radiolucency where the component parts of the maxillary sinuses should be clearly visible. Only an outline of the more highly calcified bone present in the peripheral rims of the orbital cavity, the malar bone and portions of the alveolar process are distinctly visible in the x-ray. Contrast is revealed in the dense and massively formed bone, comprising homogenous and uniformed structure.

The ill effects of malformed bone are not limited to the jaws and accessory sinuses but are often a source of physical problems. Lurie[15] found it possible to use only 1,129 of 2,100 radiographs for his study of bone-age determination in white children ranging in age from 2½ to 19 years as revealed in the radiograph. The radiographs that were not usable were from children suffering from various endocrine disorders or from marked nutritional disturbances.

Most of the collection of the specimens here dealt with is now the property and on exhibit at the National Museum-Smithsonian Institute, Washington, D. C.

Received for publication, August 30, 1951; revised by author, May 7, 1952.

CAUSES OF VARIATIONS AND MORBIDITY

The same contributing factors are generally regarded to be the causes of cellular insufficiency in bone formation and of physical aberrations such as face-jaw disharmonies. These are: (1) Heredity, a progeny from a union of diverse and contrasting physical types or the mating of physical defectives; (2) adverse environmental conditions; (3) nutrition, undernourishment, and malnourishment; inadequate, deficient, or unwholesome diet; (4) endocrinopathies, dysfunctions of the pituitary and thyroid glands culminating in the known face-jaw deformities; (5) mechanical (thumb-sucking) and other habits during early childhood which are considered as causes of malrelations of the jaws, protusion of the upper jaw, or malocclusion of the teeth. Except for infrequent congenital abnormalities, these are the potentials generally regarded as directly affecting skeletal development between birth and maturity.

Developmental disharmonies in the skeleton, even from recognized causes, are not necessarily specific in every instance. Further, we may expect to find the prevailing diversities well established long before the body is subjected to clinical scrutiny, i.e., in the prenatal shaping of the body as revealed in this inquiry.

OBJECTIVES AND METHOD OF COROLLARY STUDY

The objectives in assembling and studying the embryo-fetal material were: (a) to obtain a fairly representative cross section of physical morphologic variants prevailing in modern Europeans before the body has experienced the impacts of postnatal vicissitudes; (b) to reconstruct, as nearly as possible, the chronologic increment of skeletal growth from the earliest discernible formative period in the cleared embryo to approximately the six-month-old fetus when the soft tissues approach the limit for possible or satisfactory clearing; (c) to study by direct observation normal and anomalous conditions in the prenatal skeleton, especially those of the jaws and face that are distinctly visible in "cleared and stained" specimens and (d) to present as a corollary study evidence that will establish existing approximating or corresponding similarities whether normal or defective as they occur before and after birth.

One hundred and twenty-five human embryos and fetuses were assembled for this purpose.* These came from white patients in city and private hospital wards. Ages of the specimens were provided by attending hospital personnel. Ninety per cent of the identifiable specimens collected were males. The size and weight of the specimens differed considerably in their respective ages according to information obtained. The specimens used for clearing were reasonably established as between six weeks to six lunar months by their size and developmental states. Heads of 22 babies from full term to three months of age were x-rayed.

The specimens were arranged in glass jars containing 80 per cent glycerin and 20 per cent distilled water for the fetus and 60 per cent glycerin or less

*The Schultze method of clearing was followed in the preparation of the specimens, e.g., KOH and alizarin red S dye and glycerin.

for the embryos, according to gradations in size. Completely cleared specimens, in which the calcified bone was impregnated with the alizarin, distinctly revealed all details of the developing skeletons. The smallest cleared specimen (6 weeks [?], 10 to 12 mm.), when examined with the aid of a magnifying glass, disclosed a transparent membranous nonstained sack of the head and what appeared as an outline of collagenous substance and colored calcifying portions of the developing skeleton. The stained clavicle, mandibular arch, and minutely shaped eye balls were plainly visible. Minute particles of colored buds forming the tips of fingers and toes were faintly visible.

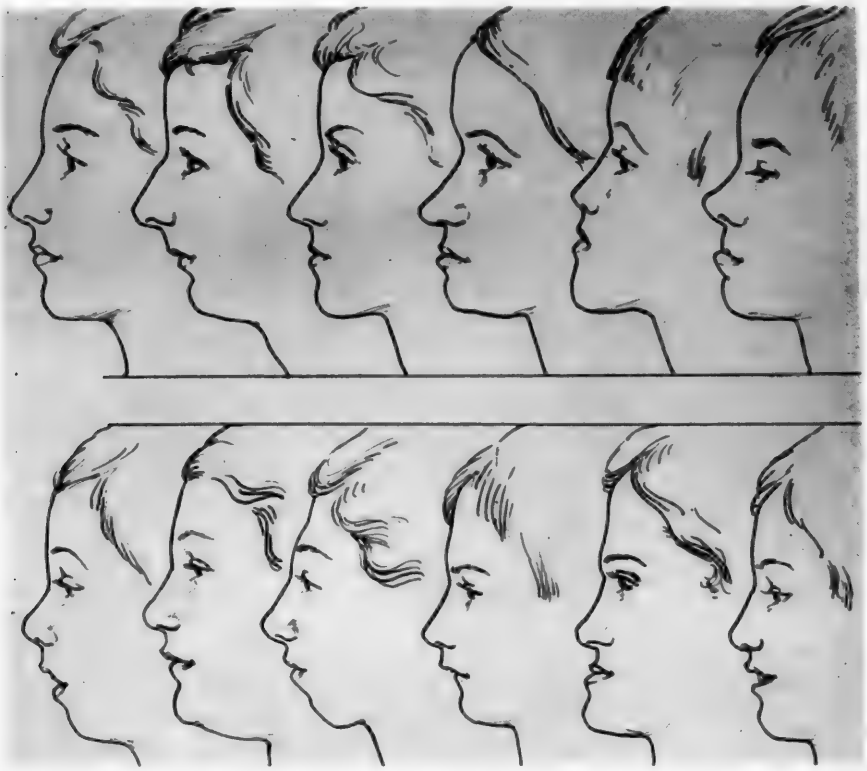


Fig. 1.—Normal and abnormal configurations of the face; marked variations in the structural pattern of the jaws; normal and disproportions in shape and size of opposing jaws in early youth (from photographs drawn to scale).

PROCESS OF OSSIFICATION REVEALED IN CLEARED SPECIMENS

The visible difference in the ossification of the component parts of the cranium in the process of transformation from a membranous sack to calcifying bone is best seen in specimens from 7 to 12 weeks. At ten weeks, the specimens disclose the proportional development of the forming face, notably the mandibular, maxillary, and nasal framework. The maxillary portion consists of the palate, alveolar border, malar process, zygoma, orbit and orbitonasal structures.

The cranial vault, though still a membranous sack, shows component segments that are beginning to calcify with the formation of a fine trabecular meshwork. The progress of growth and development of the skeleton is thereby recorded through the fetal stages up to the fifth lunar month when the soft tissues of the specimens can be cleared to translucency.*

Shaping of the Jaws.—The mandible, which already has an advanced start over the maxilla, continues to develop by depositing more of the newly formed bone along the posterior border of the ascending ramus, which rises at first at an obtuse angle, and later vertically forming a "V" shaped pattern for the formation of the condylo-coronoid process. During eight to ten weeks' period a corresponding and compensating growth is observed in the maxilla. The borders of the alveolar processes of both jaws meet in a normal state in the more advanced specimens, but the opposite is true where there are indications of a tendency toward protrusion of either the upper or lower jaws.

CONTRASTING MORPHOLOGIC VARIATIONS

The body and ramus of the mandible assume distinct angular variations. The variations are discernible in specimens of sixteen weeks and over which include the wide angle, obtuse angle and near right angle types. The shapes of the ascending ramus vary to include the broad upright type, some long and medium types and long and narrow types. The lower borders of the body of the mandible vary from a straight line that terminates in a curvature at the symphysis, to concave surfaces between the chin and the angular point of the ramus. Also the depth and width of the mandible vary among corresponding size specimens, some being of heavier and deeper types than others (Figs. 2 and 5). From the appearance of the structural morphology of the jaws in each specimen of this collection there can be no doubt of the existence of a predetermined pattern for the growth and shaping of distinct individual morphologic characteristics before birth.

Variations in the structural pattern of the jaws and the component segments of the cranium are manifest in the fetuses regardless of their corresponding size and age. The likeness or unlikeness of the head and face in shape, size, and configuration is repeatedly evident without indication of a trend toward uniformity in skeletal formation during the early stages of body growth and development. Also distinctly visible are existing differences in their heavier or lighter amounts of bone laid down in the forming jaws and component segments of the face and cranium. These contrasts are indicated by the heavier or lighter impregnation of the bone by the alizarin dye.

Ossification of the skull is markedly manifest in the 8- to 10-week-old embryo, first forming the occipital, and followed by the parietal, temporal, and frontal bones that exhibit a fine trabecular meshwork. This formation of the trabecular meshwork does not follow the same range of developmental distribution or uniformity of pattern in each specimen, nor in each specimen of the same size or age does the ossification process assume the same rate of growth or include the same proportions of calcified bone laid down in the respective cranial

*Clearing of specimens approaching sixth month is difficult or impossible unless all soft tissue is removed in addition to evisceration and decerebration.

segments. In some specimens of corresponding size the trabecular formation appears more sparse and markedly underdeveloped, especially for the frontal bone, while in others the meshwork is densely concentrated and covers wider areas of the membranous sack. This tendency toward structural variations is similarly manifest during the later periods of approximately four to six lunar months when, in addition, union of the craniofacial segments does not occur at corresponding times for specimens of the same size and age. While segments in some of the more mature fetuses are nearly closed and united, widely separated areas still prevail among others (Fig. 3).

Further differences were noted on piercing or cutting the cranial bones of the more mature fetuses. The bones in some specimens are more resistant to such treatment than others, a finding supported by x-ray examination.

A. *B.*

Fig. 2.—Five-month-old male fetuses. *A*, Larger and sturdier skeletal frame; well-shaped and fully proportioned configuration of the face, deep, sturdy, and slightly protruding mandible. Note the heavy growth of eye brows. *B*, Extremely short, shallow, and generally underdeveloped mandible—also a retarded development of mid-face and delayed shaping of the upper third of the face.

X-RAY VS. ALIZARIN IN BONE DETERMINATION

The use of the x-ray on fetuses of four months and over, to check on whether dense regions of absorbed alizarin indicate more massive bone deposition, brought surprising results. It was found that in the apparently heavily calcified bones, as indicated by the deeper staining by the dye in the craniofacial and paranasal bone structure but not in the jaws of some specimens, only a faint and hazy outline of the configuration of the head was recorded on the x-ray films. The jaw bones usually show a greater degree of calcification even though the rest of the craniofacial bones are barely visible. Some specimens which were similar in size and uniformly stained, but not as intensely impreg-

nated with the dye, appeared more homogenous in the x-ray and not unlike the bone picture of the skull seen in early infancy. Radiographs were made directly and not through the glass jar and the glycerin and the specimens had been dyed together in the same solution.

Some question arises as to the inconsistent relationship between alizarin staining of bone and the radiopacity of the developing bone. Possibly, the disparity in the staining of the bone could be due to varied chemical consistency in the calcium deposited or that the processes of calcification and ossification are not similar. Clinically, it is not questioned that the behavior to normal or abnormal stresses of mature bone differs in different individuals. It may, therefore, be postulated that this differently formed bone has its origin during embryo-fetal life.

Admittedly the circumstances under which these embryo-fetal bodies thrived and developed, or what caused life to be terminated when it did, can only be conjectured. This is not to deny, however, that similar divergencies and abnormalities continue to exist among the living. Deviations, alterations and skeletal deformities of the face that occur in the growing child or young adult are generally attributed to ill-nourishment, infantile ailments, the effects of the dysfunctioning endocrine system, and thumb sucking. Perhaps these factors may have some influence in the ultimate shaping of the jaws in the growing child. Similarly, modifications in the shaping of the craniofacial features may follow between the late fetal and full-term periods. Nevertheless, it is significant that irregularities in the jaw relationship that are present at term and thereafter also may be present in the fetus of 4 to 6 months. The visible tendencies toward protruding maxillae or prognathous mandibles as they occur at this state of development would therefore pose this question: Would these irregularities change to normal conditions were life continued? This question is particularly pertinent since such abnormalities and deformities of the jaws, as they prevail among the living, are also present in the fully developed fetus.

The like and the unlike characteristics are reflected by an array of diversely shaped malposed jaws, or narrowness of the maxillary and mandibular arches. These are clearly visible when present in the larger fetuses (4-6 lunar months). The more normal specimens exhibit alveolar ridge-to-ridge apposition or apparent normal jaw relationship.* The familiar clinical picture present in the living takes added significance by the contrasting evidence observed, for example, in the two male fetuses—estimated age approximately 5 months (Fig. 2).

For approximately the same body, length of the fetus, the shapes and sizes of the component parts of each skeleton differ. They, however, exhibit correspondingly proportionate sizes or hands and feet that are in relation with the thickness (appearance, not measured) of the long and flat bones of which the face and jaw bones are the representative part of the respective skeletons.

*For the study of the arch pattern of the jaws, a considerable number of heads were removed from larger cleared specimens, thus providing a full and unobstructed basilar view of the structural anatomy.

Fig. 3.—Variations in bone formation of the craniofacial and jaw structures, and diversity in the closure of the cranial sutures. *A*, Estimated 6-month-old fetus measuring 24 cm. "crown-heel," the largest in the collection. Note that it has not totally cleared after prolonged submersion in KOH, and 5 years' submersion in 80 per cent glycerin solution. Despite the developmental age and otherwise normal appearance, the cranial segments are ununited. *B* and *C*, Five-month-old fetuses, respectively. Note the contrasting appearance in the completion and the wide chasm in one, and almost total closure of the cranial segments in the other. *D*, Photograph. *E* and *F*, x-rays of 4-month-old fetuses, respectively. Note advanced union of segments which also reveals a densely normal trabecular structure of the entire cranium. *F*, Massively ossified jaws for so early a period of body development. For size and age of this fetus, the jaws and particularly the mandible appeared to be the heaviest and deepest of any specimen in this collection. Note: Only the x-ray will disclose actual ossification and is not comparable with the stained dimensional areas of apparent calcification.

FACTORS AND EVIDENCE ASSOCIATED WITH SKELETAL MORBIDITY

The skeletal overgrowth with enlargement of the lower jaw, hands, and feet, often associated with hyperpituitarism, may be observed in some of the specimens in this collection (Fig. 3). The influence of the ductless glands as a determining factor between physical and physiologic constancy and morbidity can not be discussed here except to cite briefly some pertinent facts of exemplary value. The function or dysfunction of the respective glands, manifest clinically in the living youth and adult, cannot, without speculation, apply to similar manifestations observed in the developing fetus. Individuals affected by glandular disorders during preadolescent periods may exhibit positive symptoms including retarded stature, yet the craniofacial skeleton may remain unaltered.

Animal experimentation in the fields of endocrinology, nutrition, and genetics has resulted in similar skeletal changes produced by different approaches to the problem. Todd and Wharton[22] produced changes in the skull growth and jaw defects in sheep through thyroid deficiency. A rachitogenic diet, fed to breeding rats by Warkany and Nelson,[23] resulted in extremely severe congenital deformities, including receding jaws noted externally in 81 of the 93 abnormal newborn. In the majority of the newborn rats thus affected the condition was so marked that the large part of the tongue protruded from the mouth. Similar morbidity prevailed throughout the bodies of these rats. Disharmonies and abnormalities of the jaws as well as other skeletal defects resulting from cross breeding of types of dogs have been amply demonstrated by Stockard[20] and by Phillips.[11] Prognathism in Jersey strain "Bulldog Head" cattle has been discussed by Becker and Arnold.[1] Acromegaly was produced experimentally in dogs by Benedict, Putnam, and Teel.[17] Overgrowth of the body, including the mandible and tongue, followed the injection of anterior pituitary extract, while retarded or arrested growth was obtained in thyroidectomized and hypophysectomized dogs. This correlating experimental evidence will substantiate the potential factors associated with skeletal aberrations outlined earlier.

No abstruse explanation is necessary to correlate the existing similarities in the biologic potentials of man and beast; what is true in one is usually true in the other. What explanations may be had for the existing extremes in disproportions of size between upper and lower jaws other than the possible human blending of contrasting physical types? A massive, large, deep, and protruding mandible may articulate inadequately with the markedly undersized maxilla, only the opposing molar teeth making partial contact. Such disharmonies are manifestly more conspicuous in the female. A variety of abnormalities and diverse traits for which there is no reasonable basis for association with glandular disorders are present in the human dental organism. Anomalies such as congenitally missing, displaced, and malposed teeth are observed, as well as marked variations in the pattern, shape, and size of the teeth in the mouth of the same person.[18]

The effects of malnourishment or undernourishment are obviously the same before or after birth. Prenatal body growth and development may suffer as

a result of maternal malnourishment or other physiologic dysfunctions. It is doubtful, however, whether such nutritional deficiency affects skeletal form or morphogenesis. This fact became evident during the process of clearing the specimens. A number of smaller specimens were devoid of the usual fat tissue; "mere skin and bone" would best describe them. Most of such specimens did not tolerate 1 per cent KOH and had to be destroyed. However, despite their emaciated condition, these specimens did not exhibit signs of skeletal deformity, nor anomalies comparable with results obtained experimentally on animals.

One theory receiving serious attention among many clinicians is the maxillary deformity attributed to the child's habit of thumb-sucking. The familiar constricted palate or protruding incisors lend valid reasons for such belief. Nevertheless, this habit is not restricted to children of Europeans. It is equally shared, if not enjoyed by the Negro, American Indian and no doubt children of other primitives and even the young chimpanzee. Yet, seldom are these deformities observable in the children of primitive origin, nor was such evidence visible in the two thumb-sucking chimpanzees I have seen; one 14 months old, obtained from its natural habitat, was at the Denis Zoological Garden (Dania, Florida) and the other, 7 years old, was born and reared at the Yerkes Laboratory (Orange Park, Florida).

There is good reason to believe that deformed maxillas in European children may be due to ill- and sparsely-formed bone that yields to thumb pressure and stretching during early childhood. However, the fact that faciomaxillary protrusion is exhibited in the larger fetuses and is further confirmed in the x-ray at birth and for several months thereafter, tends to reduce the validity of such assumption. A condition that may some time produce asymmetry in the shape of the palate is an enlarged buccal fat pad (Fig. 4). Clinch, according to Hellman,[10] reported the following: "Of a group consisting of 400 babies, at birth, the mandible of 74 or 18.5 per cent were too far back in their relationship to the maxillae."

The usual array of diversities were impressively apparent in the x-rays of 22 baby heads. The craniofacial and jaw bones in some of the specimens were of uniformly dense consistency, while in others the variations were from intermediate to an extremely contrasting type. Many of these were of sparse and porous ossification and unusually thin cranial vaults. The latter types made it exceedingly difficult, and even impossible, to section sagittally through the head as intended. In many instances, the outer skin held the cranial vault intact, while in others even the skin offered little support to the "eggshell-like" bone.

GROWTH AND DEVELOPMENT OF THE FACE

That which has been related here concerning the diverse and contrasting internal parts of the skeleton exhibited in the dyed bone, applies to the extreme features of the face that fit the head. Although growth of the face begins and continues from the lower third, first at auriculonasion line, then at the mid-face and finally by nasal frontal height, the ratio and increment of growth between the maxilla and the frontal bone do not follow uniformly in the embryo-fetal

development. These contradistinctive facial features are clearly outlined in profile after the fourth lunar month and indicate that growth of the human face does not follow and conform to an established anatomic pattern. From the appearance of these faces, it is hard to visualize a prescribed norm process of growth into which the upper or lower portion of the face must fit.

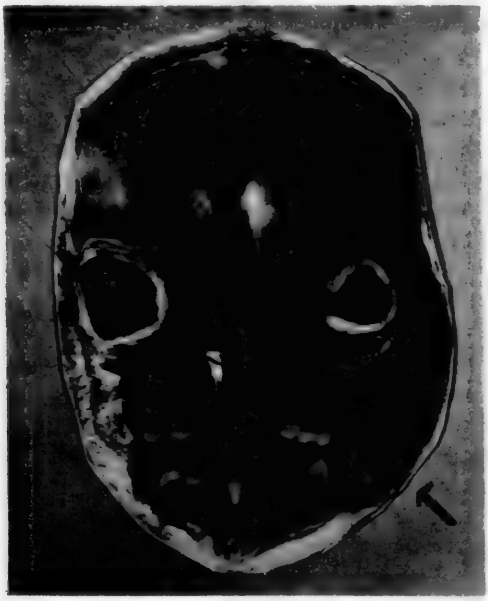


Fig. 4.—Sagittal section, anterior portion of the face of a nearly full-term baby; congenital overgrowth of buccal fat pads depressing maxillary wall, sinus, and palate with resultant deformity of right side of face.

So pronounced are the contrasts in proportion of the growing faces of either upper face height, midline or lower portions (as exhibited in each specimen) that it would rule out the possibility of a standardized process of growth and shaping of the craniofacial features. While in most instances the upper height of the face (frontal nasal) constitutes about two thirds of the full length of the face, there are other examples where fetuses of approximate size and perhaps the same age display what may be regarded as normally proportioned facial distributions (Fig. 5). Added prominence is given these diversified facial features by the presence of or lack of densely calcified bone formation of the paranasal region, the maxilla, and the mandible. All of these are distinctly brought out by the colored bone in the fully cleared specimens.

SOFT TISSUE REACTIONS TO CHEMICALS AND LIQUIDS

Variations and contrasts also are present in the soft tissue as in bone. When more than one specimen of either embryo or fetus was placed in the same vessel for dehydration, action of the alcohol was not identical in all specimens. Some specimens dehydrated relatively rapidly and completely (de-

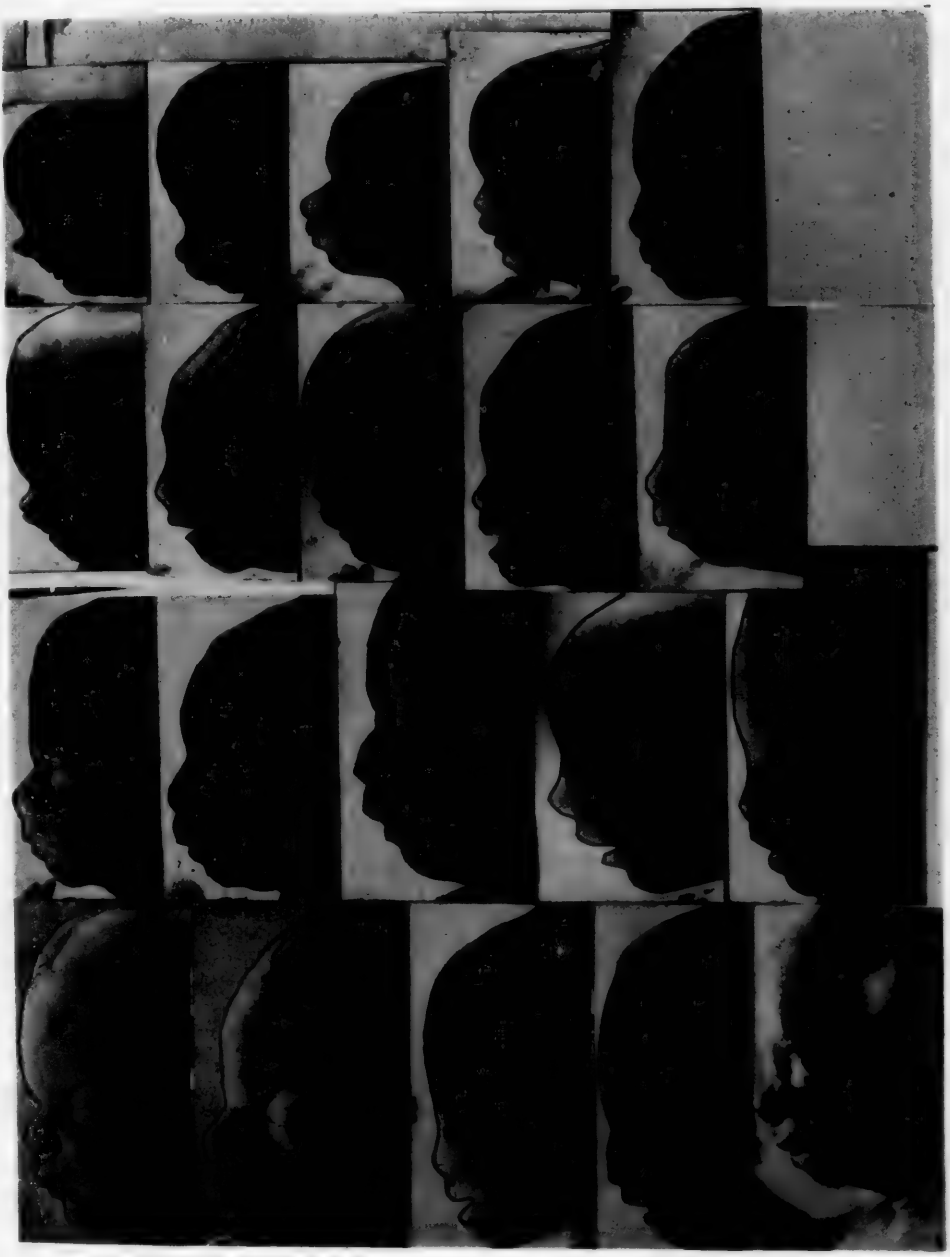


Fig. 5.—Growth and shaping of the face in the fetus. The first fetus in this group is our smallest specimen, approximately 14 weeks of age. The remaining range from approximately 16 to 24 with the exception of the fourth from the last which is over 6 months old.

pending upon the size) while others seemed to retain the body water for a longer time. The same tendency was observed when the specimens were placed in alkali (KOH-1 per cent) solution after dehydration. Some absorbed more of the alkali water and appeared like inflated dolls. In these, the soft tissue tended to become translucent more rapidly. This type remains bloated, takes up alizarin dye, and clears in the glycerin rapidly. When cleared, the soft tissues resemble a clear waxlike substance in which the colored bone stands out prominently. Other specimens do not exhibit the same tendency toward this translucency. Regardless of the size of the specimen and the fact that all were treated alike, there was marked difference in the appearance of the soft tissues. In some, the soft tissue appears more dense, resembling thicker muscular tissue. It appears that some of the smaller specimens (embryos) do not have the same tolerance for glycerin mixed with equal parts of distilled water. This is evidenced by a collapse or sagittal folding of the membranous sack of the head, from the frontal to occipital region, and the thoracic-abdominal wall, shortly after they are placed in a higher concentration of glycerin. Shrinking and shriveling of the skin occur in some of the larger specimens when placed in 80 per cent glycerin. This same proportion of glycerin and water also seems beyond tolerance of some of the eviscerated specimens which rise to the surface. Other specimens settle to the bottom of the jar containing the same solution.

SUMMARY

1. Sufficient evidence has been collected from the study of 125 embryos and fetuses to indicate that characteristics exhibited in these specimens bear close resemblance to physical differences present in the living. The differences included variations in the process of ossification of the component parts of the cranium, face, and jaws, in the shape and proportions of the developed jaws, and in the relationship of upper and lower jaws.

2. On the basis of the evidence presented, the indications are that morphogenesis of the skeletal structure and the shaping of the individual follow an inherently established pattern during embryo-fetal life. Disharmonies in skeletal growth and development occurring between birth and maturity may result from a multiplicity of factors.

3. The trend toward frequent facio-jaw deformities in civilized populations is especially evident in most Europeans where random matings are common between diverse physical types often including physically defective.

4. From observations cited in the discussion it appears doubtful whether undernourishment may result in alteration in physical morphology of the craniofacial and jaw structures during prenatal life, except perhaps for the effects of hypocalcemia.

Foremost, I am deeply indebted and grateful to my colleagues, friends and hospital personnel who untiringly provided me with specimens. I am equally indebted to Dr. Samuel Brown and Dr. Archie Fine who have generously and gratuitously given their time and material doing the x-ray work. To Dr. Arthur Mirsky and the May Institute for Medical Research (Jewish Hospital, Cincinnati, Ohio) for providing laboratory facilities. Thanks are due Drs. Elbert B. Ruth, College of Medicine, Cincinnati, Ohio; Linden F. Edwards and Hamilton G. B. Robinson, Ohio State University, for their help, guidance and comment.

REFERENCES

1. Becker, R. B., and Arnold, P. T. D.: "Bulldog Head" Cattle, *J. Heredity* **40**: 282, 1949.
2. Broadbent, H. B.: Ontogenetic Development of Occlusion, Philadelphia, 1941, Univ. of Pennsylvania Press.
3. Brash, J. E.: Growth of the Jaws, Normal and Abnormal in Health and Disease, Dental Board of United Kingdom 1924 (No. 485 lectures).
4. Brodie, A. G.: On the Growth Pattern of the Human Head: From Third Month to Eighth Year of Life, *Am. J. Anat.* **68**: 209, 1941.
5. Faucet, E.: The Development of Bones Around the Mouth, Dental Board of United Kingdom, 1924.
6. Englebach, W.: Endocrine Medicine, Springfield, Ill., 1932, Charles C Thomas.
7. Goldzieher: Abnormalities of Facial Development Due to Endocrines, *D. Cosmos* **77**: 733, 1935.
8. Grollman, Arthur: Essentials of Endocrinology, Philadelphia, 1941, J. B. Lippincott Co.
9. Hrdlicka, A.: The Causes of Malocclusion, *D. Cosmos* **64**: 489, 1922.
10. Hellman, M.: Changes in the Human Face Brought About by Development: *Internat. J. Orthodont.* **13**: 475, 1927; *Idem:* The Face in Its Development Career, *D. Cosmos* **77**: 685, 777, 1935; *Idem:* Development of Occlusion, Philadelphia, 1941, University of Pennsylvania Press.
11. Phillips, J. McL.: "Pig Jaws" in Cocker Spaniels, *J. Heredity* **36**: 6, 1945.
12. Keibel, F., and Mall, F. P.: Manual of Human Embryology, Philadelphia and London, 1912, J. B. Lippincott Co.
13. Keith, A., and Campion, G. C.: Contribution to the Mechanism of Growth of the Human Face, *D. Record* (London) **40**: 11, 1922.
14. Krogman, W. M.: Racial and Individual Variation in Certain Facio-Dental Relationships, *J. D. Res.* **14**: 277, 1934.
15. Lurie, L. A., Levy, S., and Lurie, M. L.: Determination of Bone Age in Children, Pediatrics **23**: 131, 1943.
16. Noback, C. R.: Some Gross Structural and Quantitative Aspects of the Developmental Anatomy of the Human Embryonic, Fetal & Circumnatal Skeleton, *Anat. Rec.* **87**: 29, 1943.
17. Benedict, E. B. B., Putnam, T. J., and Teel, H. M.: Studies in Acromegaly. VIII. *Arch. Surg.* **18**: 1708, 1929.
18. Rabkin, S.: Major Factors Influencing Changes in the Human Dentition, *J. Med.* **24**: 235, 1943.
19. Schour, I., and Massler, M.: Endocrines and Dentistry Council of Dental Therapeutics, *J. A. D. A.* **30**: 595, 763 and 943, 1943.
20. Stockard, C. R.: The Genetics & Endocrine Basis for Differences in Form and Behavior, Philadelphia, 1941; *Idem:* Physical Basis of Personality, Norton & Co., 1931.
21. Thompson, J. R., and Brodie, A. G.: Factors in the Position of the Mandible Jaws, *J. A. D. A.* **29**: 925, 1942.
22. Todd, T. W., Wharton, R. E., and Todd, A. W.: *Am. J. Anat.* **63**: 37, 1938.
23. Warkany, J., and Nelson, R. C.: Skeleton Abnormalities in the Offspring of Rats Reared on Deficient Diets, *Anat. Rec.* **79**: 83, 1941.
24. Weinmann, J. P., and Sicher, H.: Bone and Bones, St. Louis, 1947, The C. V. Mosby Co.

THERMAL INJURY TO NORMAL DOG TEETH: IN VIVO MEASUREMENTS OF PULP TEMPERATURE INCREASES AND THEIR EFFECT ON THE PULP TISSUE

V. F. LISANTI* AND H. A. ZANDER**

Tufts College Dental School, Boston, Mass., and University of Minnesota School of Dentistry, Minneapolis, Minn.

THE reaction of the dental pulp to heat has been a matter of conjecture for years, with no controlled histologic research. The subject becomes more important with the ever increasing use of self-curing resin filling materials which produce heat during polymerization.

While there are numerous reports concerning temperature measurements resulting from operative procedures, these have been made in vitro. The purpose of this investigation is twofold: (1) to determine the reaction of the normal dental pulp to temperatures which might be created by operative procedures or by filling materials. (2) to measure the elevations in temperature of the pulp tissue in vivo resulting from the application of heat.

Review of the Literature.—Since the advent of the motor driven hand piece, resulting temperature elevations have been a topic of concern. Bodecker,[1, 2] Bronner,[3] Jeserich,[4] and others[5-8] measured the rise in temperature within the tooth. More recently, Henschel,[9-10] and Peyton and Vaughn[11] have made contributions to our knowledge of the factors involved in heat production under controlled experimental conditions. Some of these investigators have reported temperatures up to 744.8° F. on the cutting surfaces of stone and burrs during abusive procedures. The following factors are generally acknowledged to influence thermal changes: (1) size, shape and the composition of the burr or stone, (2) speed of burr or stone, (3) amount and direction of pressure on cutting instrument, (4) amount of moisture in the field of operation, (5) length of time in continuous contact with the tissue, (6) type of tissue being cut, enamel or dentin, and (7) distance between the burr and thermocouple (in measurement work).

While there is only limited information concerning the direct resinous filling materials, Wolcott, Paffenbarger, and Schoonover[12] have demonstrated that, in the largest possible restoration, such as a jacket crown, temperatures as high as 212° F. were created at the dentin-resin junction. A temperature of 133° F. was recorded in the pulp chamber. However, these were in vitro experiments.

Though silicate and zinc phosphate cements produce heat while setting, the temperature rise is low when compared to that of the direct resinous filling materials.[12]

The effects of thermal changes on skin and subcutaneous tissues have been studied.[13-16] A local increase in temperature above the normal produces a state

This investigation was supported by a Research Grant from the Division of Research Grants and Fellowships of the National Institute of Health, U. S. Public Health Service.

Received for publication, Jan. 19, 1952.

Read at the Twenty-ninth General Meeting of the International Association for Dental Research, French Lick, Ind., March 17 to 18, 1951 (J. D. Res. 30: 448, 1951).

of hyperemia and edema. This is an indication of increased capillary permeability. A temperature of 101.2° F. applied for six hours on experimental animals caused irreversible changes in the epidermal cells.[13] For each degree centigrade rise in surface temperature between 44 and 51° C., the time required to produce a similar injury was reduced by approximately one half. Further studies with temperatures ranging from 44° C. to 1000 ° C. for one second to seven hours, disclosed that 24 to 48 hours was required to detect microscopically the line of demarcation between reversible and irreversible thermal injury.

The effect of thermal irritation on formative dental tissue was studied by passing a stream of water heated to 113° F. over an exposed tooth bud. This resulted in hyperemia, fibroid degeneration of the enamel organ, and changes in the odontoblastic layer. Increasing the water temperature from 127.4° F. to 158° F. resulted in edema, extravasation of red blood cells, dentinal degeneration, lack of enamel calcification, cellular destruction, an increase in the number of inflammatory cells, osseous replacement of pulpal tissue, and extrusion from the alveolus of the formed portion of the tooth.[17]

Much emphasis has been placed on the histopathologic observations of extracted teeth where histories have indicated thermal trauma. Little has been reported of the superimposition of other factors which influenced the final observation. Such factors as the original carious lesion, extent of cavity preparation, type of filling material used, and most important of all, the original condition of the dental pulp prior to treatment, have not been controlled or evaluated.

While there are many excellent reports concerning the reaction of the dental pulp to filling materials and drugs,[18-23] there is no information available relative to the effect of measured temperatures on the dental pulp.

EXPERIMENTAL PROCEDURE

Elevations in temperature were simulated by a device designed to apply heat to the floor of a previously prepared cavity. This device consisted of a copper thermode machined to the diameter of a No. 560 fissure burr. It was mounted on the heating element of a soldering iron. The temperature at the tip was controlled by a variac whose electrical power source (from a 110 A.C. line) was stabilized by a General Electric voltage stabilizer (Fig. 1).

The temperature increase within the pulp tissue was measured by means of a thermocouple (end area .072 mm.^{2}) connected through a cold end junction to a millivoltmeter. Its range was calibrated from 70° F. to 600° F. This was inserted into the pulp chamber by means of a hole drilled through the lingual opposite the cavity preparation. In order to align the heat source in the cavity preparation with the thermocouple inside the pulp, a threaded yoke was used. (Fig. 2.) One end of the yoke held an insulated guide for the thermode. The other end served to guide the thermocouple through an insulated sleeve into the pulp chamber in the same axis as the thermode.

Since it was impossible to obtain intact histopathologic sections after inserting the thermocouple into the vital pulp, the investigation was divided into two parts. One part was to determine in vivo the temperature rise in the pulp tissue through various thicknesses of dentin resulting from heat applied to the floor of the cavity preparation. The other part was to observe the histopathologic changes resulting from the application of the same temperature.

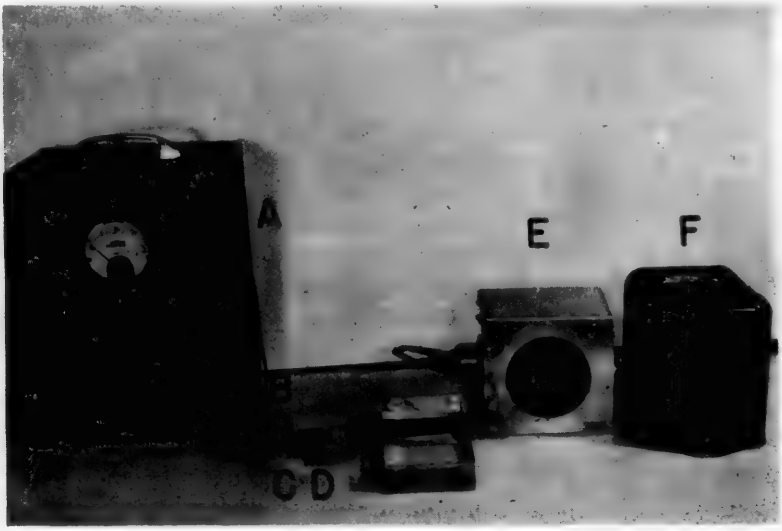


Fig. 1.—Assembly for heat application and measurement.
A—millivolt meter and cold end junction box
B—yoke
C—tooth
D—thermode
E—variac
F—voltage stabilizer

Gingival third cavities were prepared in the upper first and second incisors of five young, healthy dogs. An air stream was directed at the burr during the cavity preparation to minimize heat production. Extreme care was taken to eliminate undue pressure and friction while the burr was in contact with the tooth structure. The yoke was attached to the tooth in a buccolingual position (Fig. 1). A small hole was carefully cut into the lingual aspect of the pulp chamber with a 33½ inverted cone burr, using the lingual hole in the lingual section of the yoke as a guide. The thermocouple was inserted through the lingual guide into the pulp chamber to contact the pulpal aspect of the dentin in which the cavity had been prepared. The thermode heated to the required temperature was inserted into the labial guide contacting the cavity floor (Fig. 2). Readings from the millivoltmeter were taken every five seconds for one minute. Teeth from this part of the experiment were removed immediately, cut longitudinally through the mesiodistal plane and the thickness of the remaining dentin between the cavity and pulp measured with a micrometer. The same procedure was used for each of the five temperatures studied (125°, 150°, 200°, 300°, 600° F.).

Gingival third cavities were prepared in fourteen young, healthy dogs as described previously. The thermode heated to the required temperature was placed into the center of the cavity and held in contact with the floor of the cavity for varying lengths of time. The temperatures and the duration of application used are listed in Table I. Teeth in which cavities were prepared, but to which heat was not applied, served as a control. All cavities were filled with zinc oxide eugenol, due to its acceptance as a bland filling material. One hundred sixty-eight anterior teeth were used. The teeth were extracted at intervals of 4 hours, 24 hours, 48 hours, 1 week, 1 month, and 2 months after thermal injury.

The extracted teeth were ground in a longitudinal plane, fixed in Zenker formol, decalcified in 5 per cent nitric acid, prepared for celloidin sections and stained with hemotoxylin and eosin.

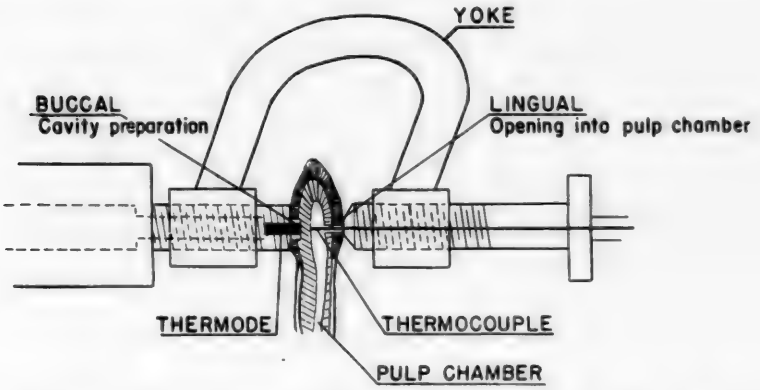


Fig. 2.—Diagram showing a cross sectional view of the thermode contacting the floor of the cavity preparation, with the thermocouple opposite the thermode in the pulp chamber.

RESULTS

The temperature increases found in the pulp tissue opposite the dentin tubules against which heat was applied, are shown in Table II. The thickness of dentin between pulp and cavity preparation varied from 0.81 to 1.52 mm. The same ranges of thickness were seen in the teeth prepared for histologic section. Therefore, the results of the histologic study may be assumed to be due to the specific increases in the temperature of the pulp tissue proper recorded in this table.

Four hours after introduction of temperatures ranging from 125 to 600° F. a separation of the odontoblastic layer from the dentin can be seen. The degree of separation is proportional to the temperature applied. There is a slight separation of the odontoblastic layer opposite the tubules corresponding to the deepest portion of the cavity in teeth extracted four hours after cavity preparation. This is the maximal irritation observed in the control specimens to which no heat was applied. (Fig. 3.) When a tooth to which no heat was

TABLE I

TEMPERATURE DEGREES F.	DURATION OF TEMPERATURE SECONDS
125	5
150	10, 20, 60
200	60
300	5, 10, 20
600	10-20

applied and its cavity preparation was filled with zinc oxide eugenol, was extracted after a month, no irritation was evident (Fig. 4). This rules out the possibility of operative procedures being responsible for the irritation observed in the pulp of teeth to which heat was applied.

When a temperature of 150° F. is applied for ten seconds, the pulp four hours later shows a greater separation of the odontoblastic layer, edema, hyperemia, and an influx of inflammatory cells (Fig. 5).

The effect of applying 200° F. for one minute is shown in Fig. 6. Four hours after heat application, the odontoblastic layer is destroyed opposite the point at which the thermode was applied. An acute inflammatory reaction is already present. One month later, the inflammation has subsided, the continuity of the odontoblastic layer has been restored, and a layer of irregular dentin has been deposited opposite the injured tubules (Fig. 7).

The effect of applying 300° F. for ten seconds is shown in Figs. 8-11. After 24 hours, there is a destruction of the odontoblastic layer and edema (Fig. 8). Forty-eight hours after cavity preparation and heat injury, there is fibroblastic activity around the area of pulp damage (Fig. 9). One week later, the area of injury approaches normal histology except for a thin area of cells in the odontoblastic layer (Fig. 10). Two months after heat injury a large amount of irregular dentin is seen opposite the cavity preparation (Fig. 11).

TABLE II

TEMPERATURE INTRODUCED (° F.)	NO. OF TEETH	THICKNESS OF DENTIN (MM.)	TEMPERATURE RISE IN THE PULP (EXPOSURE TIME)	
			5 SEC. (° F.)	1 MIN. (° F.)
125° ± 3°	4	1.10	3.0	13.8
		1.20	3.1	14.0
		.90	3.5	14.2
		.88	3.5	14.0
150° ± 3°	4	1.33	5.5	19.7
		1.21	5.7	20.1
		.92	5.0	20.0
		.81	5.2	20.1
200° ± 5°	4	1.52	10.1	25.2
		1.24	11.3	26.2
		.95	11.4	27.3
		.91	11.2	26.8
300° ± 10°	4	1.14	22.7	56.2
		.99	22.8	57.0
		1.23	21.8	55.9
		1.43	21.0	54.0
600° ± 10°	4	1.82	43.4	83.2
		1.34	44.6	86.0
		1.12	43.5	87.3
		.93	42.7	91.5

The application of 600° F. for ten seconds resulted in extensive destruction and blister formation in the pulp (Fig. 12). This reaction extends below the coronal portion of the pulp. However, after two months, even this severe injury heals with irregular dentin formation (Fig. 13).

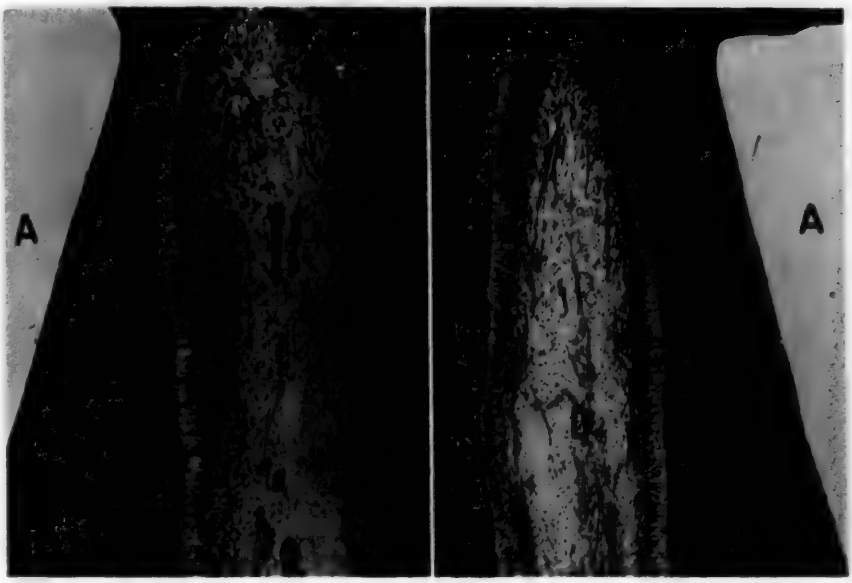


Fig. 3. Fig. 4.

Fig. 3.—Four hours after cavity preparation and placement of zinc oxide and eugenol.
A—cavity preparation
B—dentin
C—separation of odontoblasts from dentin
D—normal pulp

Fig. 4.—One month after cavity preparation and zinc oxide and eugenol restoration.
A—cavity preparation
B—dentin
C—odontoblastic layer
D—normal pulp

DISCUSSION

The experiments measuring the temperature rise of the pulp tissue show that a tooth in vivo is capable of dissipating heat rather efficiently. This is in part due to the action of the dentin, since less elevation was observed with thicker layers of dentin between the cavity preparation and the pulp (Table II). Henschel[9] has calculated that the temperature of a burr cutting dentin may go as high as 270° F. However, this was not a direct measurement. Probably, higher temperatures are reached during abusive operative procedures. The highest temperature used in this experiment, 600°F., probably represents the highest reached by a burr under most conditions. Fortunately, heat dissipating mechanisms appear to be operating which reduce the temperature increase of the pulp to a fraction of the temperature applied. The dentin itself acts as an effective insulating medium, bearing out the results of a previous in vitro study which showed dentin to have a low thermal conductivity.[24]

The histopathologic portion of this study indicates that the normal dental pulp of healthy dogs is capable of recovery from heat injury. The stages of the recovery process are as follows: (1) walling off of the area of injury, (2) the removal of necrotic tissue, (3) the replacement of the damaged tissue, (4) the deposition of irregular dentin opposite the irritated tubules.

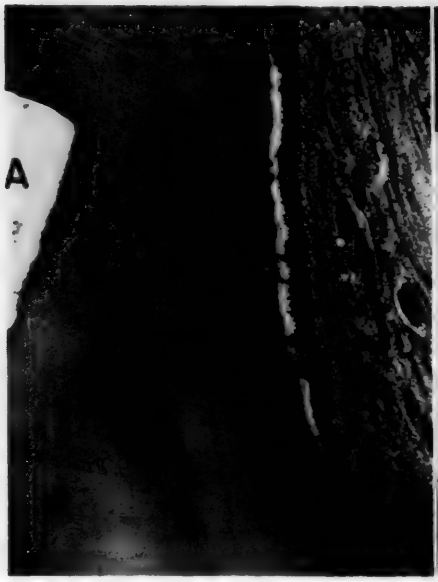


Fig. 5.

Fig. 6.

Fig. 5.—Four hours after the application of 150° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—separation of odontoblastic layer
D—hyperemia
E—inflammatory cells

Fig. 6.—Four hours after the application of 200° F. for one minute and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—destroyed odontoblastic layer with inflammatory cells
D—edematous pulp

The temperature ranges utilized in this study include those reached by filling materials and operative procedures, according to available reports. Ele-

---

Fig. 7.—One month after the application of 200° F. for one minute and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—irregular dentin
D—predentin
E—regenerating odontoblasts
F—normal pulp

Fig. 8.—Twenty-four hours after the application of 300° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—destroyed odontoblastic layer
D—area of coagulation necrosis

Fig. 9.—Forty-eight hours after the application of 300° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—area of damage
D—fibroblastic activity walling off damaged area

Fig. 10.—One week after the application of 300° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—area of healing

Fig. 7.

Fig. 8.

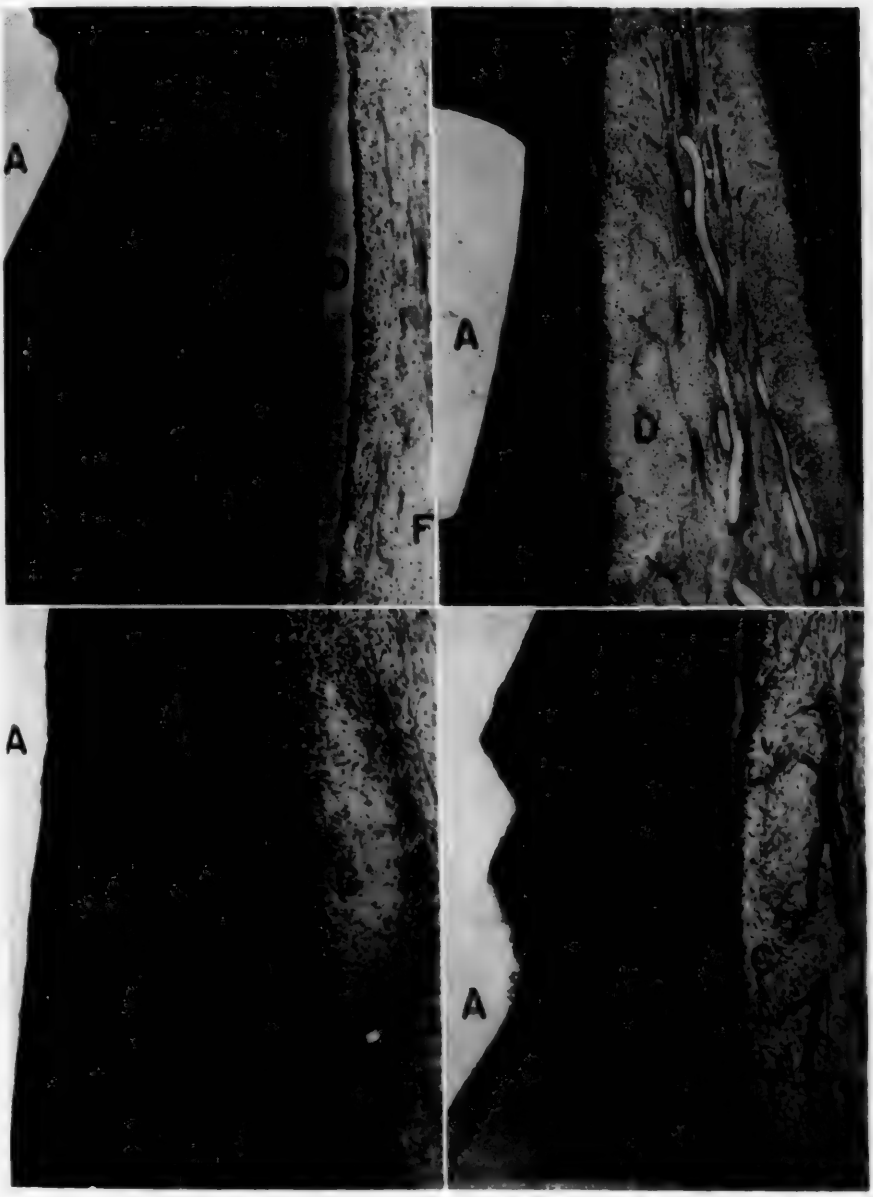


Fig. 9.

Fig. 10.

(*For legends, see opposite page.*)

vations in temperature increase the severity of the pulpal injury; however, pulp healing resulted regardless of the temperature applied. No pulp deaths were observed in this study.

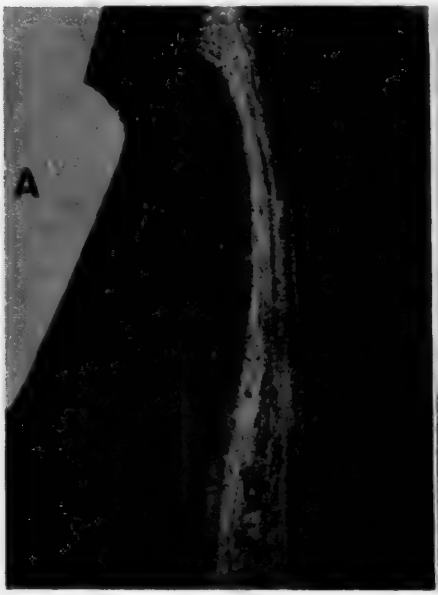


Fig. 11.

Fig. 12.

Fig. 11.—Two months after the application of 300° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—irregular dentin in previously damaged area
D—normal pulp

Fig. 12.—Fours hours after the application of 600° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—blister and formation
D—compressed pulp tissue

Since previous studies have shown a close correlation between the reaction of human and dog pulps, it may be presumed that the normal human dental pulp will react similarly. While restorative dentistry is primarily concerned with carious teeth, it is impossible to determine with accuracy any degree of pulp pathology clinically. Therefore, the expected response of the human dental pulp would depend in part upon the degree of disease already present when treatment is instituted. Repair or healing after injury would be influenced by the ability of various cells to differentiate and the extent of tissue damage. This is dependent in part upon the depth of the lesion and preparation.

SUMMARY AND CONCLUSION

Cavity preparations were made in 168 sound teeth of healthy dogs. Equipment was designed to apply measured temperatures to the floor of the cavity

preparation and to determine the resultant increase in temperature of the pulp tissue proper. Histologic sections were prepared to show the effect of heat on the pulp after varying periods of time.

The temperature increase of the pulp tissue proper is a fraction of the temperature applied to the floor of the cavity preparation. The greater the bulk of dentin between the pulp and cavity floor, the less the temperature increases in the pulp. Dentin appears to be an effective insulating material.

Normal dog pulps react to temperature applications of 125° to 600° F. by alterations in the odontoblastic layer, blister formation, and varying inflammatory changes. All pulps showed healing after thermal injury regardless of the temperature applied. The normal dental pulp appears to be able to withstand the harmful effects of temperature increases resulting from normal operative procedures.

Fig. 13.—Two months after the application of 600° F. for ten seconds and placement of zinc oxide and eugenol.

A—cavity preparation
B—dentin
C—irregular dentin formation in areas of blister formation (see Fig. 12)

REFERENCES

1. Bodecker, C. F.: Practical Dental Histology, *D. Cosmos* **70**: 893, 1928.
2. Bodecker, C. F.: Ill Effects of Heat Caused by Rapid Operative Technic, *J. A. D. A.* **26**: 527, 1939.
3. Bronner, F. J.: Some Studies on Pulp and Dentin Reactions, *D. Cosmos* **74**: 535, 1932.
4. Jeserich, P. H.: Factors Necessary to Minimize Thermal Changes in Tooth Structure from Operative Procedures, *N. Y. J. Dent.* **5**: 275, 1933.
5. Willis, H. E., and Warner, H. K.: Heat in Cavity Preparation, *Australian J. Dent.* **44**: 62, 1940.
6. Walsh, J. P., and Symmons, H. F.: A Comparison of the Heat Production and Mechanical Efficiency of Diamond Instruments, Stones, and Burrs, at 3,000 and 60,000 rpm, *New Zealand D. J.* **45**: 28, 1949.
7. Anderson, D. J., and Van Praagh, G.: Preliminary Investigation of the Temperature Produced in Burring, *Brit. D. J.* **73**: 62, 1942.
8. Wilson, G. W.: Year Book of Dentistry, Chicago, 1939, Chicago Year Book Publishers, Inc., p. 276.

9. Henschel, C. J.: Heat Impact of Revolving Instruments on Vital Dentin Tubules, *J. D. Res.* **22**: 323, 1943.
10. Henschel, C. J.: The Friction of Revolving Steel Burrs, *J. A. D. A.* **31**: 895, 1944.
11. Peyton, F. A., and Vaughn, R. C.: Thermal Changes Developed During the Cutting of Tooth Tissue, *Fortnightly Rev.*, Oct. 15, 1950.
12. Wolcott, R. B., Paffenbarger, G. C., and Schoonover, J. C.: Direct Resinous Filling Materials: Temperature Rise During Polymerization, *J. A. D. A.* **42**: 253, 1951.
13. Moritz, A. R., and Henriques, F. C., Jr.: Studies of Thermal Injury II, *Am. J. Path.* **23**: 695, 1947.
Moritz, A. R.: Studies of Thermal Injury III, *Am. J. Path.* **23**: 915, 1947.
14. Hudack, S., and McMaster, P. D.: The Gradient of Permeability of the Skin Vessels Influenced by Heat, Cold, and Light, *J. Exper. Med.* **55**: 431, 1932.
15. McMaster, P. D., and Hudack, S.: Induced Alterations in the Permeability of the Lymphatic Capillary, *J. Exper. Med.* **56**: 239, 1932.
16. Leach, E. H., Peters, R. A., and Rossiter, R. J.: Experimental Thermal Burns Especially the Moderate Temperature Burn, *Quart. J. Exper. Physiol.* **32**: 67, 1943.
17. deAprile, E. C.: *Brit. D. J.* **74**: 352, 1943.
18. Manley, E. B.: Investigations Into the Early Effects of Various Filling Materials on the Human Pulp, *D. Rec.* **62**: 1, 1942.
19. Shroff, F. R.: Effects of Filling Materials on the Dental Pulp, *New Zealand D. J.* **42**: 99, 145, 1946.
20. Zander, H. A.: The Reaction of Dental Pulps to Silicate Cements, *J. A. D. A.* **33**: 1612, 1946.
21. Glass, R. L., and Zander, H. A.: Pulp Healing, *J. D. Res.* **28**: 97, 1949.
22. Gurley, W. B., and Van Huysen, G.: Histologic Changes in the Teeth of Dogs Following Preparation of Cavities of Various Depths and Their Exposure to Oral Fluids, *J. A. D. A.* **26**: 87, 1939.
23. Fish, E. W.: The Reaction of the Dentinal Pulp to Peripheral Injury of the Dentine, *Brit. D. J.* **52**: 338, 1931.
24. Lisanti, V. F., and Zander, H. A.: Thermal Conductivity of Dentin, *J. D. Res.* **29**: 493, 1950.

THE EFFECT OF SINGLE DOSES OF IRRADIATION UPON THE TISSUES OF THE UPPER RAT INCISOR

H. MEDAK, M. WEINREB, H. SICHER, J. P. WEINMANN, AND I. SCHOUR

From the Departments of Histology and Oral Pathology, College of Dentistry, and Department of Radiology, College of Medicine University of Illinois, and Department of Anatomy, Loyola University School of Dentistry, Chicago, Ill.

MEDAK, Schour, and Klauber[11] and Weinreb[20] studied the effects of single localized doses of x-ray irradiation upon the rate of eruption of the upper rat incisor. Doses of 3,000 r. and 4,000 r. caused a marked depression of the rate of eruption during the first two weeks. Later a complete cessation of eruption resulted.

The present study deals with the histologic changes of the upper incisors of the rat and their supporting tissues, following the same method of irradiation as described in the previous report.

REVIEW OF THE LITERATURE

Many of the studies on the effects of x-ray irradiation on the dental structures were limited to clinical observations (Gotthard,[8] Stark,[18] Jüngling,[9] del Regato,[5] and Bruce[3]).

Leist[10] was the first to investigate histologically the effects of irradiation on the continuously growing rat incisor. He exposed eight rats to a dosage of 15 Holzknecht units at various intervals. These rats showed the same histologic picture: the calcified dentin was abnormally thin and the predentin was rather thick and lined by degenerated odontoblasts. Corresponding to the number of exposures, niches occurred on the pulpal surface of the dentin, indicating temporary interruption of dentin formation. The enamel showed no changes. From experiments on dogs, which were irradiated with 30 to 40 Holzknecht units and sacrificed after 2½ months, Leist[10] concluded that the odontoblasts were the most radiosensitive elements of the tooth. After irradiation they were fewer in number and completely disarranged, especially in the coronal portion of the growing tooth.

Smith[17] tried to determine whether repeated dental roentgenograms taken in children could have any effect on the developing teeth. He subjected five 6-day-old rats to x-ray exposures similar in intensity to those that a child might be subjected to in routine roentgenologic examination. He found no structural changes in the teeth or the jaws. With high doses he was able to reproduce some of Leist's findings.

Zerosi[21] reported on the effect of x-ray irradiation on the periodontal tissues of young dogs. He applied single doses of 800 and 900 r. to the jaws and sacrificed the animals 15 and 25 days after irradiation. He reported hyperemia, hemorrhage, and degenerative processes in the pulp, gingiva and, to a slight degree, in the periodontal membrane. The bone marrow showed moderate fibrous transformation.

This investigation was supported (in part) by a cancer training grant from the National Cancer Institute, United States Public Health Service.

Received for publication, March 12, 1952.

Burstone,[4] using a contact x-ray machine, studied the effects of x-ray irradiation on the teeth and their supporting structures of 44 mice, 2 to 28 days old. Doses of 5,000 r. damaged the ameloblasts severely, leading to a cessation of histodifferentiation of new ameloblasts. He confirmed Leist's findings that the odontoblasts are more sensitive to the irradiation than the ameloblasts. He described the formation of osteodentin which trapped the damaged odontoblasts and extended into the pulpal tissues.

Weinreb[20] studied the effect of single x-ray exposure of 4,000 r. upon the growth pattern of the upper rat incisor by means of alizarin red S injections. He found that in the pre-experimental part of the incisor dentin formation continued at a normal rate (about 16 μ daily). English and Tullis[6] observed in the developing teeth of swine which were exposed to 2,000 kv. total body x-radiation in doses of about 250 to 800 r. in air that the ameloblasts actively producing enamel matrix appeared to be sensitive, while other portions of the teeth showed minimal reaction to irradiation.

MATERIAL AND METHODS

The present investigation is based on the histologic study of the upper incisors of 59 adult white rats, varying in age from 30 to 90 days and in weight from 100 to 200 grams. The animals were irradiated with one single dose varying from 1,000 to 4,000 r. (Table I).

TABLE I

SUMMARY OF EXPERIMENTAL DATA

IRRADIATION DOSES IN r	NUMBER OF RATS	DURATION OF EXPERIMENTS
1000	12	1 to 35 days
2000	8	1 to 35 days
3000	12	1 to 56 days
4000	27	6 hours to 300 days

The method of irradiation has been described in detail in a previous report.[11] The apparatus used was a Keleket Therapy Machine, mobile unit (up to 140 kv.). The physical factors used were: 100 kv., H.V.L. of 1 mm. Al. (inherent filtration), 5 Ma., 12½ cm. t.s.d., a field of 2.5 cm. diameter; no additional filter was used. The output was 320 r. per minute in air. Care was taken in positioning to direct the central rays toward the basal end of the upper incisor.

The period of survival was from 6 hours to 43 weeks. Normal rats of similar age were used as controls. Immediately following the sacrifice the heads of the animals were fixed in 5 per cent formalin, and roentgenograms were taken. The upper incisors and the surrounding tissue were decalcified in 5 per cent nitric acid and embedded by routine method either in paraffin or in celloidin. The sections were stained with hematoxylin-eosin, by silver impregnation (Gomory[7]) and with Heidenhains modification of Mallory's connective tissue stain (Romeis[13]).

EFFECTS OF IRRADIATION WITH 1,000 r. AND 2,000 r.

Pulp.—The primary changes following irradiation with 1,000 r. are well localized to the labial zone of proliferation of the pulp of the basal end of the

incisor. As early as 48 hours after irradiation an edema can be observed in this area which is caused in part by the liquefaction necrosis of some of the cells. The edema is especially visible in silver impregnated sections in which some of the argyrophil fibers are widely separated while others are bundled together. Two days later the edema had disappeared and the fibers had regained their normal appearance and arrangement. Secondary changes were observed in the odontoblasts, at the dentinoenamel junction and in osteodentin formation.

Odontoblasts.—The transitory edema of the pulpal connective tissue caused permanent damage to the odontoblasts in the involved area. They cease to produce dentinal intercellular substance and appear much shorter than normal odontoblasts. Their regular arrangement also is lost. Furthermore, fine argyrophil circumnuclear granules, characteristic for the normal odontoblast, disappear from the damaged cells. The cessation of dentinal growth in the circumscribed area, while incisally and basally dentinogenesis proceeds normally, leads to a permanent baylike defect of the labial dentin, the dentinal niche (Figs. 1 to 6). This niche, of course, moves incisally with the progress of tooth eruption until it is finally lost.

Osteodentin.—The reaction of the pulp to the degeneration of the odontoblasts consists in the formation of osteodentin (Figs. 4, 5, 6), corresponding at first to the extent of the dentinal niche, although it may later transgress this area. The first sign of the formation of osteodentin is the appearance, on the third day, of small isolated masses of hyaline, staining pink with eosin, but best seen in sections stained with Mallory. These masses are separated from the dentin merely by the layer of degenerated odontoblasts. Into the hyaline mass are embedded Korff's fibers. At first acellular, this abnormal tissue during its growth includes cells in the second week and was therefore diagnosed as osteodentin.

The dentinal niche is at first fairly wide but is gradually narrowed by accumulation of osteodentin until it represents a narrow transverse cleft of the labial dentinal wall. Sometimes the layer of osteodentin is covered on the pulpal surface by a more nearly normal dentin.

Dentino-Enamel Junction.—The initial damage to the labial proliferative zone of the pulp has one more secondary effect. The rate of proliferation of the pulp is for a time depressed, while the epithelial enamel organ evidently continues to grow at a normal rate. In consequence of this break in the correlation of growth on the two surfaces of the dentino-enamel junction, the latter assumes a slightly wavy appearance (Figs. 3, 4, 5, 6). Furthermore, in some of the experimental animals there is a temporary disturbance of odontoblastic activity in the same area, characterized by inclusion of cells in, and irregular calcification of, a layer of dentin basally to the dentinal niche.

Two thousand r.—The findings in dentin and pulp in animals irradiated with a single dose of 2,000 r. did not differ principally from those after irradiation with 1,000 r. However, the first sign of the damage, the labial edema of the basal pulp, could be seen after 24 hours. The edema also was more severe and lasted somewhat longer. In accordance with this, the damage to the dentin and the waviness of dentino-enamel junction were enhanced (Figs. 2 and 3).

Enamel Organ.—In addition, irradiation with 2,000 r. caused some damage to the epithelial enamel organ. In some animals a shallow hypoplasia developed

(Fig. 2) corresponding to the enamel niche and incisal to the area in which the dentino-enamel junction was found to be wavy. The ameloblasts in the area of enamel hypoplasia were shorter than normal and were not quite as regularly arranged as those incisally and basally from the lesion.

EFFECTS OF IRRADIATION WITH 3,000 r. AND 4,000 r.

Irradiation of the rat incisor with single doses of 3,000 and 4,000 r. caused damages that were essentially similar. The changes were not only much more severe than those caused by lower doses but involved also a wider area. In addition to pulp, dentin and enamel, the high doses produced also damage to the periodontal membrane and the bone. The findings at different times after irradiation show clearly that a period of destruction is followed by one of regeneration, though these periods may somewhat overlap. The latter is highly variable in its final outcome.

1. Destructive Changes.—

Pulp: Twenty-four hours after irradiation a small area of edema was present in the pulp along the labial odontogenic epithelium (Fig. 7) which, in subsequent days increased in size and led to a liquefaction necrosis of pulpal connective tissue. This necrosis also involved the odontoblasts and prevented further differentiation of odontoblasts and formation of dentin. On the lingual surface of the pulp the edema started one day later and the area of edema and necrosis was smaller.

After four days the necrotic area on the labial surface extended half-way across the pulp and progressed incisally along the odontogenic epithelium. By the fifth day the necrotic area was well circumscribed and could be called a cystic cavity (Fig. 8). Eight days after irradiation the necrosis, which extended beyond the cystic cavity, had reached across the base to the most basal tip of the lingual surface. By the sixteenth day the cystic cavity, which meanwhile by its pressure had caused a partial exfoliation of the tooth, had fused with a cystic cavity that had gradually developed in the labial alveolar periosteum. The fused cysts eventually filled the whole base of the socket and were invaded by the sixth and eighth week by young connective tissue. In some instances formation of bone occurred in this granulation tissue.

Several areas of hemorrhage at the basal end became apparent as early as the fourth day (Fig. 7) and could still be observed after twenty-four days. The blood vessels of the entire pulp were dilated, filled with red blood cells, and some were thrombosed.

Dentin: While the odontoblasts in the area of edema and necrosis ceased the further formation of dentin, the older odontoblasts continued to form dentin until, in about five weeks, the pulp cavity became obliterated. The dentin formed after the fourteenth day frequently contained vascular inclusions.

The demarcation of the dentin at its basal end was more sharply outlined on the lingual than on the labial side (Fig. 8). Lingually the dentin ended sometimes abruptly, whereas the labial dentin continued for a short time as a thin layer.

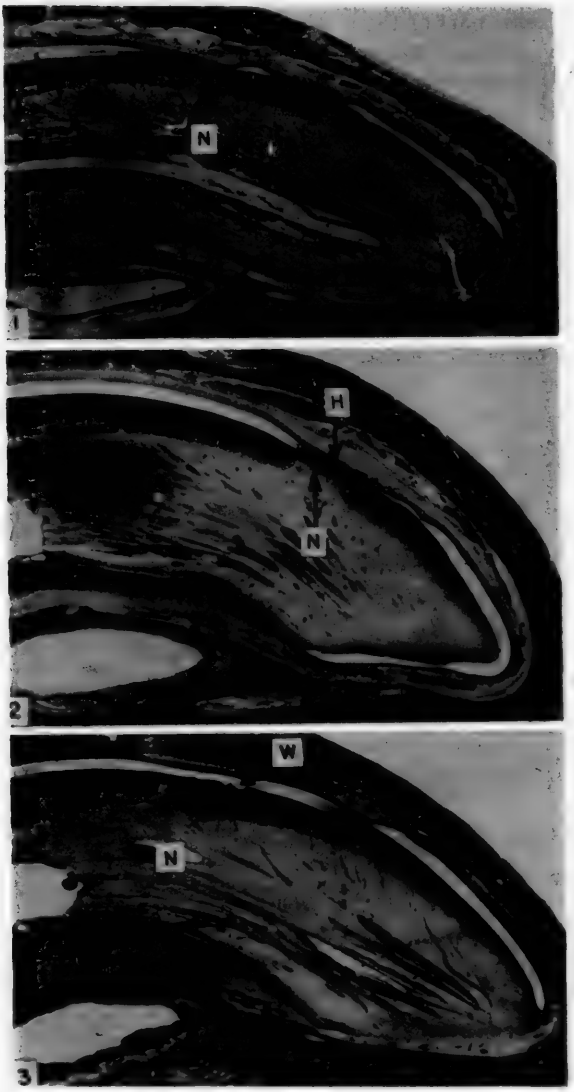


Plate I

Photomicrographs of decalcified longitudinal central sections through the basal part of upper rat incisors; hematoxylin-eosin stain.

Fig. 1.—Ten days following irradiation with 1000 r. Note the dentin hypoplasia forming the dentinal niche (N). Osteodentin has formed in the niche. The hypoplastic defect developed at the time of the irradiation and had moved forward with the eruption of the tooth.

Fig. 2.—Fourteen days following irradiation with 2000 r. Note the dentinal hypoplastic niche (N). The enamel formed since the time of the irradiation is also hypoplastic (H).

Fig. 3.—Three weeks following irradiation with 2000 r. Note the hypoplastic dentinal niche (N) and the waviness of the dentino-enamel junction (W) basally to the dentinal niche.

Osteodentin: On the second day following irradiation osteodentin could be observed in the basal part of the tooth. The first sign of osteodentin appeared in the zone of the damaged odontoblasts on the labial side. The osteodentin developed parallel to the layer of damaged dentin but remained separated from it by the layer of degenerated odontoblasts. It eventually fused with the thickened pre-experimental dentin and extended toward the pulp. Together with the osteodentin, which developed on the lingual surface and extended labially, it formed a diaphragm which narrowed the opening of the pre-experimental pulp cavity. The odontoblasts separating the layer of damaged dentin from the osteodentin became smaller and compressed as the osteodentin became bulkier.

Enamel and enamel organ: The youngest ameloblasts in the odontogenic zone, which had not yet begun to produce enamel matrix, were inhibited to such a degree that they did not, after irradiation, produce enamel matrix. The ameloblasts in the next zone, that had already produced a thin layer of enamel matrix gradually increasing in thickness incisally, continued to produce enamel matrix, but finally stopped their activity. The closer they were to the basal end the sooner their activity ceased. In other words, the younger the active ameloblasts, the less enamel matrix was formed before their function came to an end. Only the oldest matrix producing ameloblasts continued to form enamel matrix of normal thickness. As a result the basal zone of enamel matrix tapered abnormally toward the basal end (Fig. 7).

Maturation of the enamel matrix proceeded undisturbed until all matrix was completely calcified by the sixteenth day (Fig. 8). Due to the cessation of eruption, the ameloblasts did not move incisally; therefore, sixteen days following irradiation, the physiologic age change of the enamel organ into reduced enamel epithelium had proceeded noticeably toward the basal end and, at the fifth week, the entire pre-experimental organ had changed into reduced enamel epithelium.

The odontogenic epithelium which appeared normal up to the second day began to show a continuously increasing edema between the inner and outer enamel epithelium. The pulpal surface of the odontogenic epithelium was separated from the pulpal connective tissue, first by necrotic tissue and later by the cyst. Beginning on the third day, cells of the inner enamel epithelium became shorter and from the fourth day on appeared flat or low cuboidal. By this time an edema and necrosis had developed in the labial alveolar periosteum and had started to displace the odontogenic epithelium progressively toward the lingual side. By the tenth day the pressure of the cyst of the labial alveolar periosteum and the cyst in the pulp had reduced the odontogenic epithelium to a thin strand of cells, located close to the midline of the tooth (Fig. 9). When, in the third week following irradiation, the two cysts had fused, the odontogenic epithelium had disintegrated and only remnants of it could be found linguobasally to the cyst.

In some animals the tooth seemed to continue erupting and the odontogenic epithelium increased in length up to about 10 days following irradiation (Fig. 7). The odontogenic epithelium on the lingual surface also continued to proliferate at least for a certain period of time (Fig. 7). However, after the seventh

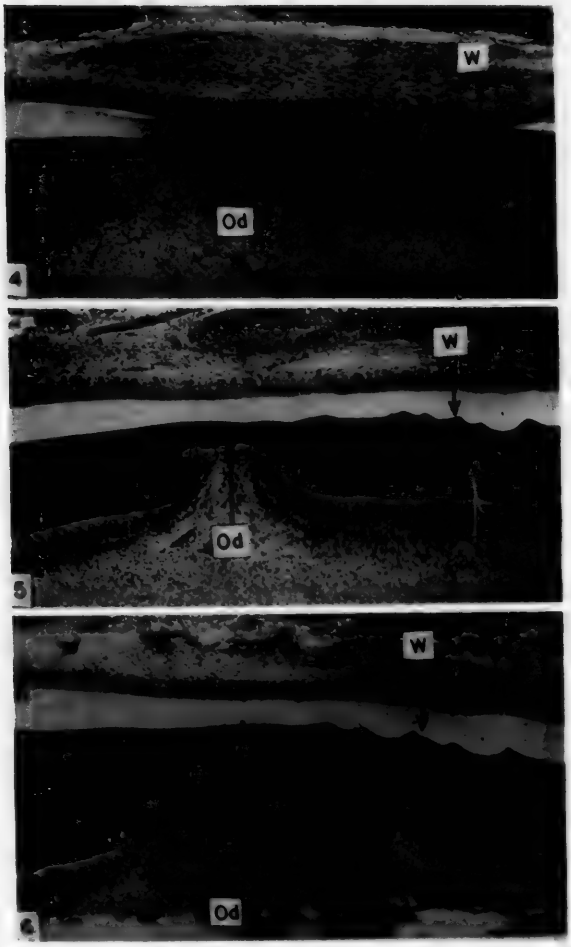


Plate II

Figs. 4 to 6.—Higher magnification of the area of dentinal niche, one, two and three weeks respectively after irradiation with 2000 r. Note the progressive accumulation of osteodentin (Od) and the waviness of dentino-enamel junction (W).

day following irradiation, the lingual odontogenic epithelium became separated from the basal end of the dentin by ingrowing connective tissue and the distance between the basal end of the dentin and the epithelium increased progressively (Fig. 9). As a result, the epithelial band gradually increased in length until only a small island of the most basal portion remained.

A small cyst developed in the lingual alveolar connective tissue adjacent to the odontogenic epithelium. This cyst (Fig. 8) was first noticed on the tenth day following irradiation and increased gradually in size. By the twelfth day it extended into the connective tissue separating the odontogenic epithelium from the basal end of the lingual dentin. This cyst too eventually became organized and blended into the connective tissue filling the base of the socket. Bone formation was observed in this region (Fig. 10).

Supporting tissues: Periodontal membrane as well as bone showed changes due to the disturbance in function of the tooth and the changed relations of the tooth to the socket.

The changes in the periodontal membrane were characterized by a gradual disappearance of the intermediate layer of the periodontal membrane and a narrowing of the periodontal membrane. The reduction in width of the periodontal membrane was first noticed eight days after irradiation, when the intermediate plexus was hardly noticeable. The fiber bundles which normally are interlaced in the intermediate plexus run in an oblique direction from the cementum covered surface of the tooth to the alveolar bone, thus simulating the arrangement of the periodontal fibers in a tooth with limited growth.

2. Regenerative Changes.—As mentioned before, all animals showed resumption of proliferation of the odontogenic epithelium, but its survival and ability to perform its functions seemed to be correlated to the extent of the labial cysts. Twelve out of twenty-seven animals sacrificed after the eighth day following irradiation showed successful regenerative processes which led to the formation of a new, though more or less distorted, incisor.

In the incisor of a rat sacrificed nine days following irradiation, an enamel organ had developed close to the labial surface of the socket, separated from the pre-experimental tooth by a layer of osteodentin (Fig. 11). A thin layer of dentin had been formed along the enamel organ. Adjacent to the dentin a well-circumscribed area of pulpal tissue could be seen characterized by densely arranged young connective tissue cells. Fourteen days following irradiation and later, the enamel organ had continued to grow and had begun to curl around the base of the bony socket (Fig. 12). After three to four weeks, the basal end of the new enamel organ had reached the lingual side of the socket.

Three animals, one sacrificed six weeks after irradiation with 3000 r. and two sacrificed ten and eleven weeks following irradiation with 4000 r., showed well-advanced regeneration of the incisor. While the newly formed dentin was fused to the pre-experimental dentin by a mass of osteodentin, and while on the lingual side close to the pre-experimental tooth irregular masses of dentin had formed, there was an increasing regularity toward the basal end of the regenerating tooth (Fig. 13). The basal end itself showed a structure and organization almost equal to that of a normal tooth. The undisturbed shape of the epithelial

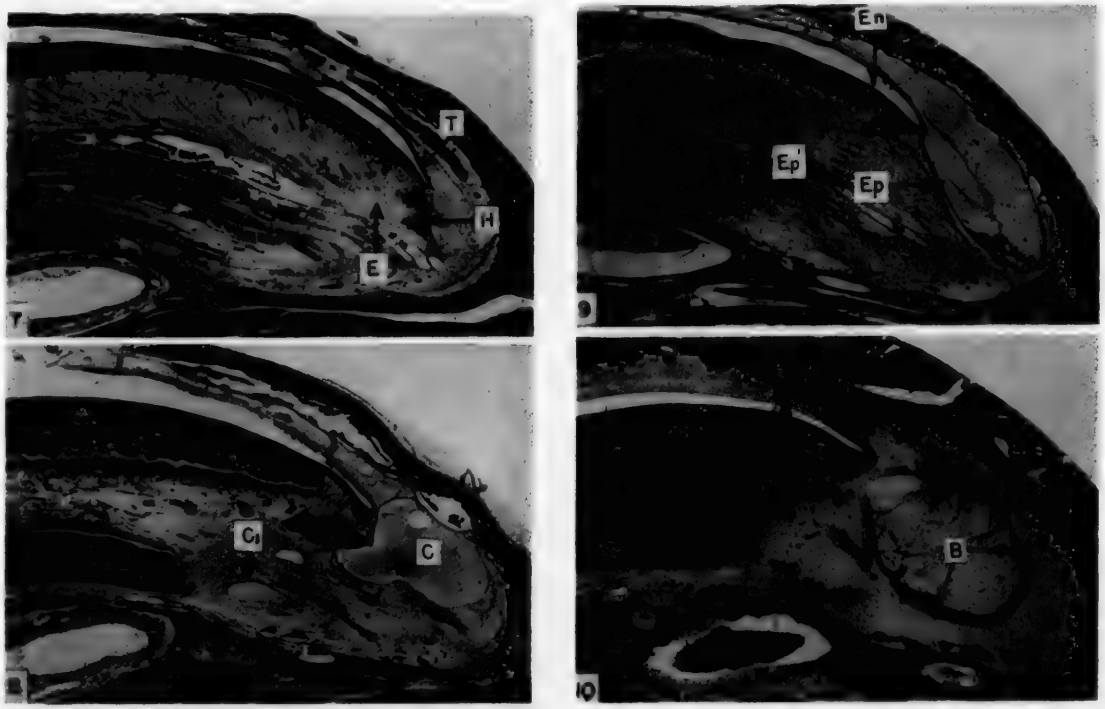


Plate III *Plate IV*

Plate III.—Photomicrographs of decalcified longitudinal central sections through the basal part of upper rat incisors six days (Fig. 7) and three weeks (Fig. 8) following irradiation with 3000 r.

Fig. 7.—Note edema (E) and hemorrhage (H) at the base of the pulp and the rapid tapering of the enamel-matrix (T).

Fig. 8.—Note large labial cyst (C) and a small lingual cyst (C_1).

Plate IV.—Photomicrographs of decalcified longitudinal central sections through the basal part of upper rat incisors 18 days (Fig. 9) and six weeks (Fig. 10) following irradiation with 4000 r.

Fig. 9.—Note the reduction of the labial odontogenic epithelium (Ep) to thin layer of low cuboidal cells, separation of lingual odontogenic epithelium (Ep^1) from basal end of pre-experimental dentin by edematous connective tissue and complete calcification of enamel matrix (En), as result of cessation of eruption.

Fig. 10.—Note base of the socket filled with connective tissue in which new bone is forming (B).

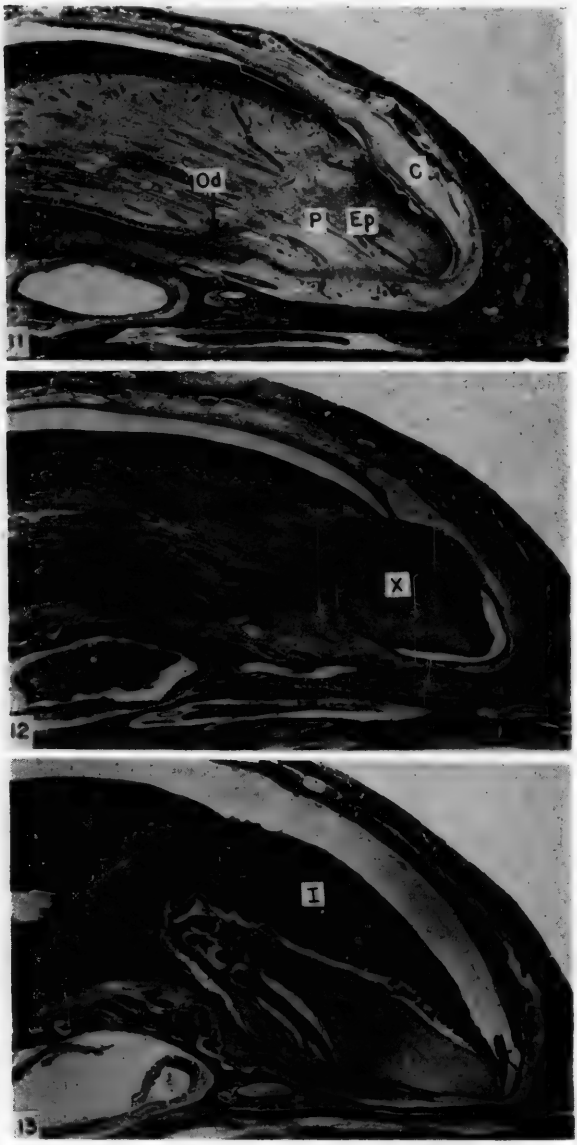


Plate V

Photomicrographs of decalcified longitudinal central sections through the basal part of upper rat incisors nine days (Fig. 11), 20 days (Fig. 12) and 11 weeks (Fig. 13) following irradiation with 4000 r.

Fig. 11.—A new odontogenic epithelium (Ep) between the arrows has formed with deposition of dentin and enamel and formation of new pulp (P). Note the cyst (C) in labial alveolar periosteum and osteodentin (Od) in pulp.

Fig. 12.—Note the resumption of the proliferation of the odontogenic epithelium and of the formation of dentin and enamel.

Fig. 13.—Note the formation of a new incisor (I), fused to the pre-experimental tooth by irregular masses of hard tissue.

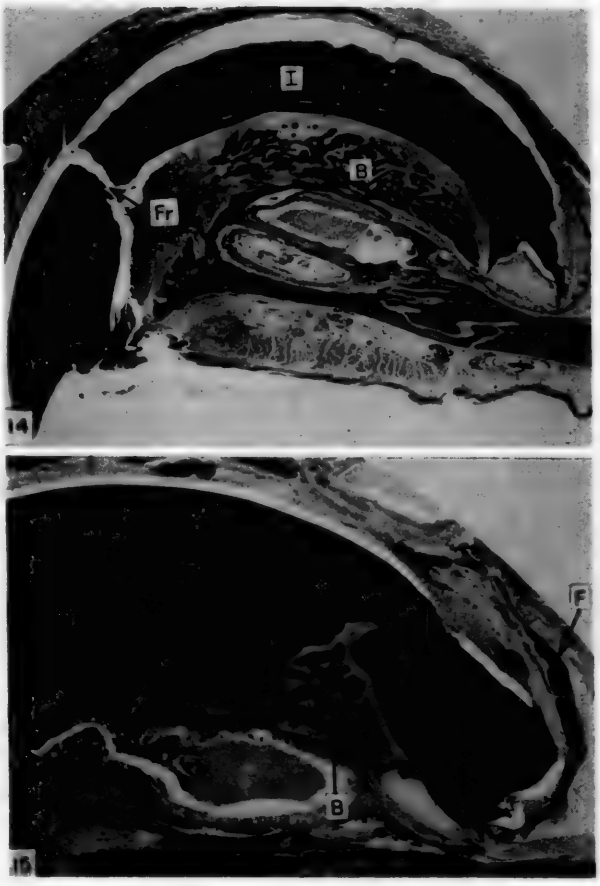


Plate VI

Photomicrographs of decalcified longitudinal central sections through the basal part of upper rat incisors 27 (Fig. 14) and 30 weeks (Fig. 15) following irradiation with 3000 r.

Fig. 14.—The regenerated tooth (I) occupies three-fourths of socket and is separated by fracture (Fr) from pre-experimental section of tooth. Pulp cavity is almost completely occluded. Functional odontogenic epithelium can be seen at basal end. Dentino-enamel junction is wavy. New bone (B) has been formed on lingual alveolar wall socket.

Fig. 15.—Regeneration was abortive. No active odontogenic epithelium is present. The fundus of the socket (F) has been rebuilt. The periodontal membrane is thin and non-functioning. Bone (B) has formed in the cleft between pre-experimental and regenerated tooth.

diaphragm, and the presence of zones of proliferation in the pulpal tissue adjacent to the diaphragm seem to indicate not only progressive growth, but in all probability progressive eruption of the regenerated tooth.

In an animal sacrificed twenty-seven weeks after irradiation (Fig. 14), the regeneration had progressed so far that the new tooth extended over three-quarters of the length of the socket. The pre-experimental incisor had "erupted" and was now separated from the regenerated tooth by a fracture. The remnants of the pre-experimental tooth, which were partly surrounded by inflammatory infiltration, had lost their organic connection with the lingual bone during the progressive exfoliation. The regenerated tooth is well curved, but has only half the diameter of a normal tooth and occupies the labial half of the socket. The lingual alveolar bone had been reinforced by spongy trabeculae, which in their entirety form a new lingual alveolar wall. The periodontal membrane on the lingual side was of fairly even width along the basal three-fourths of the regenerated tooth.

While only remnants of a pulp cavity could be seen in the incisal portion of the regenerated tooth, forming a narrow cleft close to the lingual surface, the pulp cavity was wide at the basal end occupying in longitudinal sections a triangular area. Whether this basal part of the pulp was in connection with the pulpal vestiges farther incisally, could not be ascertained.

The rather complete regeneration as described above was observed only in one animal. In one other animal sacrificed thirty weeks after irradiation (Fig. 15) the regeneration seems to have been abortive. An almost solid block of dentin was formed that nearly filled the fundic portion of the socket. The newly formed dentin was connected to the pre-experimental dentin by a callus of osteodentin. This connection was situated labially, while lingually a wedge-shaped cleft remained between pre-experimental and regenerated dentin. This cleft was filled by spongy bone which however did not seem to be fused at any point to the osteodentin or cementum covering the tooth. The movement of the pre-experimental incisor seems to have ceased entirely, as shown by the attrition reaching far beyond the gingival margin and into the socket itself. The periodontal membrane on the lingual side of the pre-experimental tooth was very narrow and did not show a functional arrangement of fibers. Both on the labial and on the lingual side of the tooth osteoclastic destruction of much of the alveolar bone could be observed. The inflammation that occurred in consequence of the abnormally deep attrition of the tooth had led to the disappearance of the anterior portion of the hard palate. The defect was filled with partly necrotic, partly inflamed tissue.

DISCUSSION

The findings on the rat incisor after irradiation with single doses of 1000, 2000, 3000, and 4000 r. confirmed and considerably extended the early findings of Leist[10] and the later findings of Burstone.[4] The evaluation of the present findings led to additional knowledge of the biology of the rat incisor or, more generally speaking, of continuously growing teeth, but also added to our understanding of tissue reaction to irradiation.

Mechanism of Tooth Eruption.—The eruption of continuously growing teeth can be defined as the visible manifestation of their growth. The "force" of eruption is the growth potential of the pulpal connective tissue. A peculiar auxiliary structure, the hammock ligament, suspends the tooth and prevents its growth into the socket. The growth of the epithelium is exactly correlated to that of the pulp (Sicher[15]).

The primary destruction of the fast growing basal end of the pulpal connective tissue by roentgen irradiation does indeed result in a deceleration and finally in a cessation of the eruption of the rat incisor (Medak, Schour, and Klauber[11]).

The fact that the x-ray irradiation with low doses resulted in a temporary injury to the pulpal connective tissue while the enamel epithelium was not affected, led to an anomaly of the outline of the tooth. Because the pulp was inhibited in its growth, the normally proliferating epithelium was thrown into folds of variable height. Waviness of the dentino-enamel junction and of the enamel surface was the effect.

A similar but more severe injury could be observed in the regenerating tooth if the pre-experimental incisor mechanically prevented the eruption of the regenerate. It is evident that the pressure built up in the confined space basally to the pre-experimental tooth will primarily prevent growth of the connective tissue of the pulp. The epithelium, however, that offers an almost linear surface to the pressure will be much less inhibited in its growth than the pulp upon whose cross-sectional surface the pressure acts. The epithelium is thrown into heavy folds and may even invade the pulp itself.

These findings are comparable to the folding of epithelium, dentino-enamel junction, and finally of dentin and enamel following hypophysectomy (Schour and Van Dyke,[14] and Sicher[15]). They are also comparable to the effect of mechanical devices that prevent attrition and eruption of the rat incisor (Arce and Erausquin[1]). The invasion of the pulp by the fast growing epithelium also reminded one of the behavior of epithelium in Vitamin A deficiency. In the latter case the break of correlation between growth potential of connective tissue and epithelium may either be caused by stimulation of the epithelium or by an unequal depression of proliferation of epithelium and connective tissue.

Periodontal Membrane.—The periodontal membrane of continuously growing teeth shows a characteristic differentiation of its principal fibers into three layers (Sicher[16]). An intermediate plexus is interposed between the alveolar fibers, anchored in the bone, and the dental fibers, anchored in the cementum. The intermediate plexus, consisting mainly of precollagenous fibers, is the site of the reconstruction of the suspensory apparatus during the rapid changes in the relation between tooth and bone.

In those teeth that have ceased to erupt after irradiation the characteristic layers of the periodontal membrane have disappeared. The principal fibers can be followed uninterrupted from bone to cementum. Their arrangement is very similar to that of teeth with limited growth.

Histodifferentiation.—According to the differentiation of enamel organ, enamel, and dentin, the normal rat incisor can be divided into six zones

(Weinmann,[19] and Pindborg[12]). The topographic relations of these zones are fairly constant, as for instance the site of enamel maturation, or the point of transition of the well differentiated enamel organ into the reduced enamel epithelium. The constancy of these points is of course the effect of a close coordination in time and space of the processes of attrition, growth, eruption and histodifferentiation of the rat incisor.

Our experimental material proves that this correlation can be disturbed in the sense that despite the inhibition or cessation of growth and eruption, the processes of histodifferentiation continue. This is the reason why in late stages the entire enamel organ has changed into the reduced enamel epithelium. For the same reason the entire enamel undergoes maturation and nothing is left of the acid insoluble enamel matrix.

These findings seem to show that the described changes are on the whole, age changes of the enamel producing organ and of the enamel itself coordinated to, but not dependent on, growth and eruption.

Appositional Growth.—The appositional growth of dentin in the normal tooth is correlated to the tempo of its growth and eruption. Inhibition of eruption however does not interfere with the process of dentinal apposition. The result is, of course, an abnormal thickening of the dentinal wall and an abnormal reduction of the volume of the pulp in a slowly erupting or non-erupting rat incisor.

Tissue Reaction to Irradiation.—The difference in susceptibility to roentgen irradiation can be viewed under four headings (Bloom[2]): the differences in sensitivity of different species of animals; individual differences within a given species; the different susceptibility of different organs; and, finally, of different cells.

The great variation in individual sensitivity is borne out by the present findings in the white rat. Success or failure of regeneration depends largely on such variations.

The differences in the susceptibility of the different cells have been explained by the supposition that more primitive cells are more highly sensitive than cells of higher differentiation, and also by the assumption that cells in mitosis are especially sensitive to the influence of the irradiation. All these assumptions are to some degree contradicted by other findings; for instance, by the resistance of spermatocytes in the testes as compared to the susceptibility of spermatogonia, though the former are practically always dividing (Meiosis).

From the results described in this report we might conclude that the cells of the connective tissue at the growing end of the incisor are more sensitive than the epithelial cells. It could also be concluded that in the connective tissue the highly differentiated odontoblasts are more sensitive than the more primitive cells of the pulpal mesenchyma.

Both these conclusions could not be entirely justified. It is true that the first injury occurs in the connective tissue and that the most striking damages are caused by irreparable injury to young odontoblasts. A closer scrutiny reveals, however, that the first sign of any damage appears to be necrosis of cells and edema in the labial proliferating zone of the pulp. It is quite possible that

the injury to the odontoblasts is not due to an inherent radiosensitivity of these cells, but that it is secondary to the circulatory damage in the neighboring tissue. The latter explanation seems to be borne out by the fact that the zone of injured odontoblasts is always restricted to the zone of pulpal damage. Especially striking is the fact that after irradiation with low doses odontoblasts of varying age are injured on the labial surface of the tooth only, while those of identical age and differentiation on the lingual side escape damage. The lasting effect (dentinal niche) of the damage to the odontoblast also is not a sign of their higher sensitivity but rather a consequence of the lack or incompleteness of regeneration or replacement of odontoblasts. In contrast, the primary damage to the pulpal mesenchyma after irradiation with low doses is soon and completely repaired.

In a comparison of the sensitivity of the epithelial enamel organ, especially the ameloblasts, and the connective tissue of the pulp, two explanations seem tenable. It is possible that the resistance of the ameloblasts is due to their higher differentiation. Against this conclusion speaks the observation that also the not yet highly differentiated cells of the epithelial diaphragm do not show any signs of primary damage. The second possible explanation for the greater sensitivity of the pulpal cells is the fact that the pulp grows at a rate that is a multiple of that of the epithelium. The latter explanation is strengthened by the observation that within the uniform primitive connective tissue of the pulp the area of most active growth is also the area of earliest and most severe damage by irradiation. It is a well-known fact that the labial half of the rat incisor grows faster than the lingual half and it is here that the low radiation doses cause the only visible injury. The great importance that growth rate has in determining the susceptibility of a tissue to irradiation does of course not exclude the well known inherent differences of tissues and cells. One should, however, be aware of the difficulty of analysis of an individual case. In a fast growing tissue the susceptibility induced by its growth potential may overcompensate the inherent resistance of its elements to radiation.

SUMMARY AND CONCLUSIONS

1. The investigation is based on the histologic study of the upper incisors of 59 white rats, 30 to 90 days of age, which had been irradiated with a single dose varying from 1000 to 4000 r. focused on the basal region of the incisors.

2. *Effect of Low Doses (1000 and 2000 r).*—The changes were localized to the labial part of the odontogenic zone and consisted of an edema of short duration and of a permanent injury to the odontoblasts in this zone, causing a dentin hypoplasia (dentinal niche). Following the degeneration of the odontoblasts, osteodentin was formed in the adjacent area of the pulp. A slight inhibition of the pulpal growth without correlated retardation of the epithelial proliferation led to waviness of the dentino-epithelial junction. A slight injury to the ameloblasts caused a shallow enamel hypoplasia.

3. *Effect of High Doses (3000 and 4000 r).*—The changes could be subdivided into two though slightly overlapping phases, namely, into destructive and regenerative processes.

Destructive changes: The localized severe edema in pulp and periodontal tissues at the basal end led to formation of cystic cavities that caused destruction of odontoblasts and cessation of differentiation of new odontoblasts. In the pulp great masses of osteodentin were formed. The cessation of pulpal growth and eruption caused a progressive maturation of the entire enamel matrix and a change of the entire enamel organ into reduced enamel epithelium, also a disappearance of the intermediate plexus in the periodontal membrane.

Regenerative changes: The outcome of the regenerative process depended on the amount of destruction caused by the expanding cystic cavities. Only if viable remnants of the odontogenic epithelium persisted to the time of organization of the cyst, the epithelial proliferation could initiate the formation of a new incisor. This regeneration was quite limited even under most favorable circumstances.

4. The primary damage of irradiation was located in the zone of most intensive proliferation of the fastest growing tissue of the tooth, namely, at the labial zone of proliferation of the pulpal tissue at the basal end of the incisor.

REFERENCES

1. Arce, C., and Erausquin, J.: Supresion experimental de la erupcion en los incisivos de la rata, *Revista Odontologica* **32**: 502, 1944.
2. Bloom, W.: Histopathology of Irradiation, New York, 1948, McGraw-Hill Book Co.
3. Bruce, Karl W.: The Effect of Irradiation on the Developing Dental System of the Syrian Hamster, *Oral Surg., Oral Med. and Oral Path.* **3**: 1468, 1950.
4. Burstone, M. S.: The Effect of X-Ray Irradiation Upon the Teeth and Supporting Structures of the Mouse, *J. D. Res.* **29**: 220, 1950.
5. del Regato, J. A.: Dental Lesions Observed After Roentgentherapy in Cancer of the Buccal Cavity, Pharynx and Larynx, *Am. J. Roentgenol.* **42**: 404, 1939.
6. English, J. A., and Tullis, J. L.: Oral Manifestations of Ionizing Radiation I. Oral Lesions and Effect on Developing Teeth of Swine Exposed to 2000 K.V. Total Body Irradiation. Naval Medical Research Institute, Dec. 29, 1950. Project NM 006012.-04.34.
7. Gomory, G.: Silver Impregnation of Reticulum in Paraffin Sections, *Am. J. Path.* **13**: 993, 1937.
8. Gotthard, P. P.: Ueber Zahnschaedigungen nach Roentgenbestrahlungen. Fortschr. a.d. Geb. d. Roentgenstrahlen, Hamb. 30, Kongressheft 139-142, 1922.
9. Jüngling, O.: Roentgenbehandlung Chirurgischer Krankheiten, Leipzig, 1924, Thieme.
10. Leist, M.: Ueber die Einwirkung der Roentgenstrahlen und des Radiums auf Zaehne und Kiefer, *Strahlentherapie* **24**: 268, 1926.
11. Medak, H., Schour, I., and Klauber, W. A., Jr.: The Effect of Single Doses of Irradiation Upon the Eruption of the Upper Rat Incisor, *J. D. Res.* **29**: 839, 1950.
12. Pindborg, J. J.: Den kroniske fluor-og cadmium-forgiftnings indflydelse paa den hvide rottes incisiver med saerligt henblik paa emaljeorganet. *Tandlaegebladet* **51**: 136, 1950. Suppl. No. 1.
13. Romeis, B.: Mikroskopische Technik, ed. 14, Muenchen, 1938.
14. Schour, I., and Van Dyke, H. B.: Changes in the Teeth Following Hypophysectomy. I. Changes in the Incisor of the White Rat, *Am. J. Anat.* **50**: 397, 1932.
15. Sicher, H.: Tooth Eruption: The Axial Movement of Continuously Growing Teeth, *J. D. Res.* **21**: 201, 1942.
16. Sicher, H.: Bau und Funktion des Fixations apparates der Meerschweinchenmolaren, *Ztschr. f. Stomatol.* **21**: Oct., 1923.
17. Smith, R. A.: Effects of X-rays on the Developing Teeth of Rats, *Am. J. Orthodont. and Oral Surg.* **24**: 428, 1938.
18. Stark, Ernst: Secundarschaeden in der Roentgentherapie, *Strahlentherapie* **16**: 600, 1924.
19. Weinmann, J. P.: Recovery of Ameloblasts, *J. A. D. A.* **30**: 874, 1943.
20. Weinreb, M.: The Effect of a Single Dose of X-Ray Irradiation Upon the Eruption and the Growth of Dentin in the Upper Incisor of the Albino Rat, Thesis, University of Illinois, 1949.
21. Zerosi, C.: Experimentelle Forschungen ueber die histologischen Reactionen and Veraenderungen der dentalen und peridentalen Gewebe infolge von Roentgen- und Radiumbestrahlung, *Ztschr. f. Stomatol.* **38**: 238, 1940.

CLEANING EFFECTIVENESS OF DENTIFRICES

GRANT VAN HUYSEN, D.D.S., AND THOMAS M. BOYD, A.B., D.D.S.

Indiana University School of Dentistry, Indianapolis, Ind.

MOST persons who are conscious of their teeth would like to have them free from dingy films or deposits. It is not only believed that such tooth surfaces look better, but that they give out less odor.[1] It is also known that most gingivitis is caused by local irritation and that accumulations on the teeth brought about by faulty oral hygiene constitute the greatest source of local irritation. The part that the dentifrice actually plays as an adjunct to the toothbrush in this matter of appearance and the maintenance of oral hygiene has only been partially determined.[2-5]

Phillips[6] studied the abrasiveness of dentifrices and the resulting effect on tooth luster. This study showed that smooth, lustrous tooth surfaces were produced by a sodium metaphosphate type of dentifrice. Phillips also showed that this smoothness of surface might be brought about with no appreciable abrasive effect on enamel and with the development of a minimum amount of erosion of exposed cervical dentin. Later Phillips[7] showed that surfaces of extracted teeth with a high luster accumulated fewer bacteria from contact than did surfaces with a low degree of luster.

Several serious attempts have been made to study the deposits usually found on teeth. These opinions and findings have been carefully summarized by Vallotton.[8] There are of course several different types of unsightly tooth surface accumulations recognized today. Exactly what these stains are, how they form, how they attach themselves to the teeth, and how they may be prevented from forming are questions not yet answered. Until these points are better understood, it is our opinion that there will be a need for a better method of comparing tooth cleanliness so that the effectiveness of dentifrices, brushes, and techniques can be properly evaluated. It is, therefore, the purpose of this study to compare the cleansing effect of various dentifrices upon the visible surfaces of the teeth. This has been accomplished by having dental students brush their teeth for three weeks each with several different dentifrices. Kodachrome photographic records are used to estimate the comparative effectiveness of the dentifrices. Comparisons are made on the basis of the differences in cleanliness of the tooth surfaces visible to the naked eye.

PROCEDURE

This study is divided into four parts but the same procedures were used in each section. The teeth of the subject were first thoroughly cleaned and polished with scalers and an engine-driven rubber cup with Mynol tooth-cleaning paste. A disclosing solution was used to make sure that there was no removable stain or debris remaining on the visible surfaces of the teeth. The participant

Supported in part by funds from the Pepsodent Division of Lever Bros. Co.
Received for publication, Feb. 29, 1952; revised by author, April 1, 1952.

was then given a numbered test dentifrice and a new toothbrush. After this dentifrice had been used for three weeks, the labial surfaces of the 12 anterior teeth were photographed. Each individual brushed his teeth with water just before the photograph was made so that recently added soft debris would be removed, since the study was concerned with the more adherent stains that accumulate over a three-week period and not those which might have occurred from the last meal.

A Zeiss "Ikon" 9 by 12 cm. double extension bellows camera with a No. 828 roll film and ground glass adapter was used for the photography. Two No. 2 photoflood lamps mounted 2 inches laterally to the lens furnished the light. The teeth were focused on the ground glass mounted on the camera back; this glass was then moved down out of the way as the roll film holder moved into position for exposure of the film. Kodachrome No. 828 A film was used. All photographs were taken under similar conditions in regard to time of exposure and lighting. Two exposures were made in each instance, and before each exposure was made the teeth were focused on the ground glass of the camera back.

After each dentifrice had been used for three weeks and the appearance of the teeth recorded on the Kodachrome film, the teeth of the individual were again cleaned and polished. The individual was given another one of the dentifrices in the test series to use for another three-week period. The cleansing, three-week brushing period, and photographs were repeated until all of the dentifrices of the test series were used by each of the members of the group. The photographs taken after each three-week period provided a permanent record of the amount and character of the stain which had accumulated during that period.

The Kodachrome films, after they had been processed, were marked with a code number and arranged in envelopes with the name of the individual to whom they belonged on the outside. Each envelope finally contained the sets of duplicate photographs representing each dentifrice tested for each participant in the study. The better one of each of the films was selected and arranged on a uniformly lighted background. They were then rearranged from left to right according to the cleanliness of the teeth. When the observer decided which Kodachrome transparency showed the cleanest teeth, it was placed on the extreme left of the uniformly lighted area. The Kodachrome showing the next cleanest teeth was placed to the right of this and so on until the color photographs of one individual taken after using each of the dentifrices had been arranged side by side for comparison. Where teeth showed equal cleanliness, although different dentifrices had been used, the color photographs were placed in the same position. After the Kodachrome films had been arranged in the order of cleanliness of teeth, the number of the dentifrice used for three weeks to obtain the degree of cleanliness was revealed and scored. Table I shows the frequency with which the Kodachrome records of tooth cleanliness were placed in the comparative positions. Selections of comparative cleanliness viewed in the color photographs were made independently by several observers. The summary of scores of various observers may be seen in the table.

Since all of the subjects brushed their teeth with water just before the teeth were photographed at the end of the three-week test period, the assessment of cleanliness depended upon the appearance of more or less fixed deposits such as Manley's pellicle and stained granular calculus, that is, the more adherent films (Fig. 1).

The test dentifrices* used were all toothpastes; their code number and composition are listed below:

TABLE I

CODE NO.	FORMULA TYPE OF TOOTHPASTE
13	Chalk and soap.
29	Water-soluble chloryphyllins, chalk, and synthetic detergent.
84	Tricalcium phosphate, synthetic detergent, diammonium phosphate and high urea content (12.5%).
76 and 98	Insoluble sodium metaphosphate, dicalcium phosphate and sodium lauryl sulfate.
56	Tricalcium phosphate, sodium lauryl sulfate, low urea (3%) and diammonium phosphate.
96	Dicalcium phosphate and synthetic detergent.
86	Tricalcium phosphate, sodium lauryl sulfate, diammonium phosphate and low urea content.
66	Dicalcium phosphate and sodium lauryl sulfate.

DATA ON STUDY NO. 1

In Part 1 of this study, 15 subjects used four different dentifrices, each one for a period of three weeks. The results of the study are recorded in the attached table and are described herewith. Two observers studied the color photographs taken after each three-week period the individuals used a dentifrice. Observer 1 was of the opinion that 12 of the 15 (80 per cent) of the individuals kept their teeth cleanest with toothpaste 76, which was composed of insoluble sodium metaphosphate, dicalcium phosphate, and sodium lauryl sulfate. Observer 2 was of the opinion that only 8 (53 per cent) of the group, were able to keep their teeth cleanest with toothpaste 76. Observer 1 was of the opinion that 2 (13 per cent) of the individuals kept their teeth cleanest with dentifrice 66. On the other hand, observer 2 was of the opinion that 4 (27 per cent) kept their teeth cleanest with toothpaste 66. Observer 1 believed that toothpaste 96 failed to keep any of the individuals' teeth clean, though observer 2 thought that 3 (20 per cent) of the individuals kept their teeth cleanest with toothpaste 96. Observer 1 thought that toothpaste 86 kept the teeth of 1 (7 per cent) of the group of 15 individuals, clean, whereas observer 2 believed that toothpaste 86 failed to keep any of the individuals' teeth cleanest.

When an average of the values of the observations of the two observers is made, one can see in the last line under Study No. 1 of Table II, that toothpaste 76, the one composed of insoluble metaphosphate, dicalcium phosphate and sodium lauryl sulfate, was most effective as a tooth-cleansing agent 66 per cent of the time, whereas toothpastes 66, 96, and 86 were most effective in 20, 10, and 4 per cent of the instances, respectively.

*The dentifrices 13, 29, 84, 76, 96, and 98 were bought on the open market and repackaged in numbered tubes. Dentifrices 56, 66 and 86 are new formulas. They are described above with as much information as is generally available.

DATA ON STUDY NO. 2

Part 2 of the study shows (Table II) a comparison of the effectiveness of four dentifrices used by 32 subjects. Again each toothpaste was used by each individual for a period of three weeks and Kodachrome transparencies of the results were compared to determine the comparative cleanliness of the teeth. In Table II, under Study No. 2, one can see that observations were made by five instead of two individuals. Toothpaste 76, composed of insoluble sodium metaphosphate, dicalcium phosphate and sodium lauryl sulfate, was compared with 96, composed of dicalcium phosphate and synthetic detergent; 56, composed of tricalcium phosphate, sodium lauryl sulfate, urea and diammonium phosphate; and 66, composed of dicalcium phosphate and sodium lauryl sulfate. Study 2

TABLE II

STUDY NO.	NO. OF SUBJECTS	OBSERVER	DENTIFRICE NO. AND PER CENT OF EFFECTIVENESS							
			76	%	66	%	96	%	86	%
1	15	1	12	80	2	13	0	0	1	7
		2	8	53	4	27	3	20	0	0
		Ave.	66		20		10		4	
2	32		76	%	96	%	56	%	66	%
		1	25	78	9	28	6	20	10	31
		2	18	56	14	44	12	38	9	28
		3	21	66	14	44	15	53	11	34
		4	17	53	15	47	11	34	10	31
		5	16	50	12	38	6	20	9	28
		Ave.	61		40		33		30	
3a	29		98	%	13	%	29	%	84	%
		1	16	55	13	45	10	35	5	17
		2	20	69	18	62	13	45	9	31
		3	17	59	13	45	14	48	10	35
		4	19	66	20	69	16	55	8	28
		5	25	86	9	31	8	28	4	14
		Ave.	67		50		42		25	
3b	12		98	%	13	%				
		1	10	83	5	42				
		2	7	58	6	50				
		3	9	75	5	42				
		4	9	75	3	25				
		5	9	75	7	58				
		Ave.	73		43					

Instances and per cent of times dentifrices kept individuals' teeth cleanest.

(Table II) shows the averages of the observations made by five individuals. One can see in that line that in 61 per cent of the instances, toothpaste 76 was observed to be most effective as an adjunct to the toothbrush. Since there were occasions when one or more of the dentifrices kept the teeth equally clean, and therefore could be listed together as first choice for effectiveness, the percentages listed exceed 100. This meant that some individuals could use with equal effectiveness more than one of the dentifrices in keeping their teeth clean for a three-week period. It can be observed that toothpaste 96 was on an average 40 per cent effective, toothpaste 56 was 33 per cent effective, and toothpaste 66 was 30 per cent effective in keeping the teeth clean. There is no question in viewing

this summary that again under the conditions of the experiment toothpaste 76, composed of insoluble metaphosphate, dicalcium phosphate and sodium lauryl sulfate, kept more individuals' teeth cleaner than did any of the others, namely, 96, 56 or 66.

DATA ON STUDY NO. 3A

In the third study (3a) toothpaste 98 was of the same composition as that of 76, namely, insoluble metaphosphate, dicalcium phosphate, and sodium lauryl sulfate. During the period of the experiment the investigators used numbers only. Since the observers knew nothing about the contents of the toothpastes, the selection of the numbers in the table was made on the basis of Kodachrome study only.

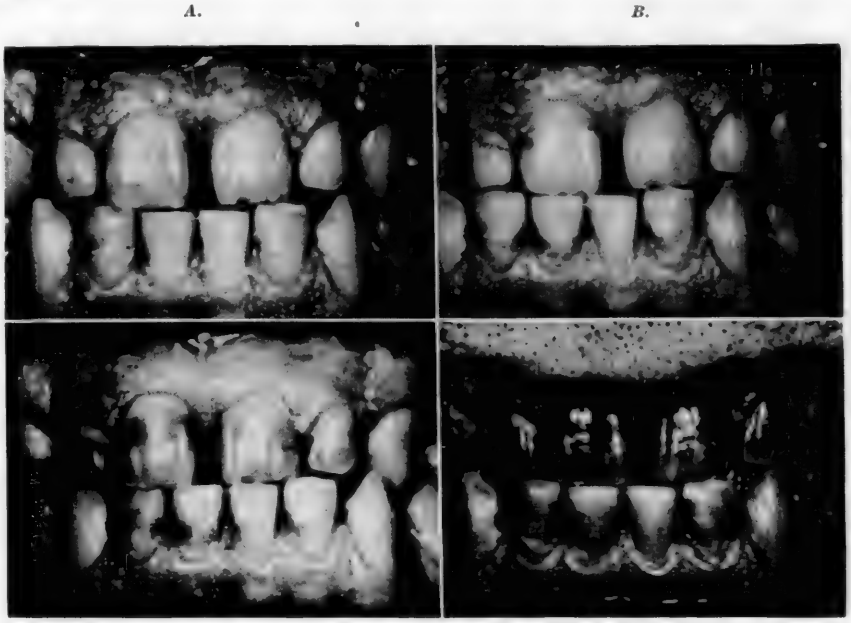


Fig. 1.—Black and white copies of a set of Kodachrome transparencies. *A*, cleanest teeth. *D*, dirtiest teeth.

Under Study 3a (Table II) one can see that toothpaste 98 was an average of 67 per cent more effective in keeping the teeth the cleanest under the conditions of the experiment. Toothpaste 13 was 50 per cent effective, toothpaste 29 was 42 per cent effective, and in an average of only 25 per cent of the times it was used, toothpaste 84 (the poorest toothpaste) was effective in keeping the teeth cleanest. The reader is again reminded that some of the participants in this study kept their teeth clean or developed the same degree of cleanness with more than one dentifrice. When more than one Kodachrome photograph showed clean teeth, the dentifrices used were recorded in the table as first choice. Where more than one photograph showed equal cleanliness but not equal to first or second position, they were listed as second or third choice, respectively.

DATA ON STUDY NO. 3B

It was decided that a part of Study 3a be repeated. A group of 12 individuals, therefore, each used dentifrices 98 and 13 for a period of three weeks. When the Kodachrome transparencies were studied, a key number was given them so that it was impossible for the observer to know which was which.

Table II shows that when the results of toothpastes 98 and 13 were compared, 98 kept the teeth cleaner in 73 per cent of the instances, whereas 13 kept the teeth as clean or cleaner in only 43 per cent of the instances in which it was used. Here again some individuals could use these two dentifrices with equal effectiveness in keeping their teeth clean for a three-week period.

DISCUSSION

The value of the method used in this study is that the observer can compare the effectiveness of the different dentifrices all at one time, and in addition the Kodachrome photographs are a permanent record that can be studied by any one at any future date. Where records are made from direct observations of the cleanliness of the teeth at the end of each of the test periods, the observer cannot go back and review slight differences not recorded on previous observations.

The variation in the frequencies of the observations made by the individual observers was studied. It is possible to show that the averages of the observations of the cleanliness of the teeth for different dentifrices differ significantly. It is therefore possible to show that the majority of a group of individuals can do a more efficient job of keeping their teeth clean with one type of dentifrice than they can with another. In this particular study, although there were some differences of opinion among observers concerning the effectiveness of the dentifrices in keeping the teeth clean during the short test interval of three weeks, in each instance the insoluble sodium metaphosphate, dicalcium phosphate, sodium lauryl sulfate dentifrice was found to be most effective for more people. The fact that the evaluation of the cleanliness of the teeth was purely subjective could account for this difference of opinion. Apparently some individuals can keep their teeth clean for a three-week period with almost any dentifrice. In the preliminary phases of this study, some individuals kept their teeth clean for three weeks with only water and a toothbrush.

It is true that the colors of the Kodachrome transparencies vary. It was demonstrated to our satisfaction, at least, that this difference occurred during processing. The observer must train himself to observe naturally occurring accumulations on the teeth and not make color differentiations. In other words, the presence or absence and the quantity of the accumulations and not the color of the debris is important in evaluating cleaning efficacy of a dentifrice.

SUMMARY

1. A method is described for comparing the cleansing effectiveness of dentifrices by the observation of permanent records by several observers.

2. A dentifrice containing insoluble sodium metaphosphate as an abrasive proved more effective when used than did those containing chalk, tricalcium phosphate, or dicalcium phosphate.

REFERENCES

1. Sulser, G. E., Lesvey, T. A., and Fosdick, L. S.: The Reduction of Breath and Mouth Odors by Means of Brushing the Teeth, *J. D. Res.* **19**: 173, 1940.
2. Fosdick, L. S.: The Reduction of the Incidence of Dental Caries, *J. A. D. A.* **40**: 133, 1950.
3. McCauley, H. B., et al.: Clinical Efficacy of Powder and Paste Dentifrices, *J. A. D. A.* **33**: 993, 1946.
4. Howitt, B. F., Fleming, W. C., and Simonton, F. V.: Study of the Effects Upon Hygiene and Microbiology of the Mouth of Various Diets, With and Without the Use of the Toothbrush, *D. Cosmos* **70**: 575, 1928.
5. Kitchin, P. C., and Robinson, H. G. B.: How Abrasive Need a Dentifrice Be? *J. D. Res.* **27**: 501, 1948.
6. Phillips, R. W., and Van Huysen, G.: The American Perfume and Essential Oil Review, January, 1948.
7. Phillips, R. W., and Schwartz, M. L.: Personal communication.
8. Vallotton, C. F.: An Acquired Pigmented Pellicle of the Enamel Surface, *J. D. Res.* **24**: 161, 1945.

ORIGIN OF ENAMEL DROPS AND CEMENTICLES IN THE TEETH OF RODENTS

VIKTORS KALNINS

Upsala, Sweden

SOMETIMES small enamel or cementum bodies are found at the side of the roots of teeth. These structures are enamel drops (or pearls) and cementicles, which are either free or may become attached to the surface of the tooth.

In the case of enamel drops, it has been assumed that the epithelial cells of the inner layer[7, 10, 11, 13, 35] or unatrophied remnant of Hertwig's sheath (Malassez's epithelial rests), provoked by some unknown factor, are able to produce enamel on the dentin of the root.[2, 7, 10, 11, 13, 35, 36, 39] Since enamel drops generally are located under the amelo-cementum junction, or under the bifurcation, it has been supposed that the development of enamel drops takes place only in the initial stage of root formation.[13] Later, the cells of the inner layer of Hertwig's sheath do not differentiate into ameloblasts and do not produce enamel.[13]

Cementicles may form through the influence of highly vital epithelial cells (scattered rest cells of Hertwig's sheath) upon the surrounding tissue of periodontium,[11] or they may develop around degenerated and "dystrophically" calcified epithelial rests of Hertwig's sheath.[1, 3, 14, 30] In both cases the further formation of cementicles takes place by true apposition of cementum[3, 11, 30] or by calcification of hyalinized connective tissue of periodontium.[3, 30] It has been assumed that cementoblasts were not necessarily involved in this process.[8, 14]

Until now studies on the structure and origin of enamel drops[4, 10, 11, 12, 18, 24, 26, 27, 29, 33, 35, 36, 39, 40, 43, 45] and cementicles[1, 3, 5, 8, 9, 11, 14, 25, 30, 31, 37, 38, 40, 41, 42] were chiefly performed on materials provided from man or animals with teeth of limited growth.[2, 13, 34, 46]

The task of the present work is to investigate whether both enamel drops and cementicles originate from the cells of enamel epithelium. For these studies rodents* were selected, because their continuously-growing teeth show all the stages of enamel and cementum development in one and the same longitudinal section. Moreover, another advantage of these teeth is the presence of enamel only on one surface while the remaining surfaces are covered with cementum. Such a structure of rodents teeth allows separate examination of developmental disturbances of enamel and of cementum.

Received for publication, October 15, 1951; revised by author, May 26, 1952.

*Data on these species guinea pigs[19, 20, 22, 23] and rabbits[21] are presented in the author's papers on vitamins and hormones. Data on rats are taken from a yet unpublished work on the effect of dehydrotachysterin (ATIO) on teeth. Moreover, the present study is completed by courtesy of other research workers with materials of their own experiments on fluorosis in guinea pigs[33] and avitaminosis A in rats.[16, 17] The material and methods of experiments are described in the above mentioned publications.

RESULTS

The findings show that enamel drops are produced from active ameloblasts which became detached from the surface of growing enamel matrix. The phenomenon of this detachment of ameloblasts depends on the particular unstable and vulnerable junction between the ameloblastic layer and the growing enamel matrix, not only post mortem,* but also intravitam (Fig. 1).

The instability of this junction intravitally is shown by observations made on our guinea pigs, which had been receiving various preparations through stomach feeding by catheter. The enamel layer was elevated from the enamel matrix. The reason for these changes was obviously the forced opening of the mouth with a wooden catch, through which the catheter was introduced (Fig. 3). Further, foldings (Fig. 4) or the detachment (Fig. 8) of the ameloblastic layer were produced by means of jarring the teeth of rats by repeatedly grinding off their crowns. It is obvious that during these manipulations the incisors of guinea pigs and rats were subjected to severe pressure which was transmitted into the formative portion of the teeth.† In both cases the formation of enamel drops (Figs. 3 and 5) could be observed. Such disturbances of enamel were not present in other untreated teeth of the same animals. No drops are formed when the detachment of the ameloblasts occurs in that portion of tooth where the enamel matrix has reached its ultimate thickness (fifth sector of Weinmann) (Figs. 4 and 8).

The examination of teeth in many normal animals showed that the detachment of ameloblasts with the subsequent formation of drops is a rare phenomenon. In scorbutic guinea pigs (Fig. 1) and in rats with avitaminosis A (Fig. 16) ameloblasts detach and enamel drops develop as a result to the folding or bending of tooth wall associated with insufficient production or hypocalcification of dentin. Enamel drops could be observed more often in connection with tooth bending in guinea pigs cured of scurvy. In these animals the bending takes place because the teeth are often displaced during the scorbutic period but in the course of healing their newly developed basal portion grows in a normal direction. Hence the two portions (the "scorbutic" and the "newly developed" one) form an angle (Fig. 5*a*). Associated with this distortion of the shape of tooth, the ameloblastic layer is stretched or folded, resulting in a disruption or a detachment of ameloblasts from the surface of enamel matrix. If, in the course of the detachment, the ameloblasts are only slightly raised from the enamel matrix, adherent drops develop (Figs. 3 and 6). If the detachment or disruption of a more or less long sector of ameloblasts layer is more marked, free enamel drops are produced (Figs. 1, 5-7, 9-14, and 16). According to the degree and extent of the detachment, the drops vary in number, size and shape.

*The instability of the ameloblast-matrix junction post mortem, due to preparation of histologic specimens, causes the regular appearance of more or less pronounced artificial space or the "gap" between the ameloblastic layer and the growing enamel matrix. If this "gap" is narrow it is regarded by some investigators as the intravital condition which is necessary for the amelogenesis. Since it is possible to make such histologic preparations where this narrow space is absent, the conclusion may be reached that this "gap" is really only an artifact (Fig. 2).

†According to Boyle, the force of mastication in the teeth of rodents normally is absorbed and dissipated only in that part of periodontium nearest to the occlusal surface of the tooth while the apical area where a constant forming of enamel takes place is protected from the functional stress.

Usually they are of a globular, oval or ellipsoid appearance. In some cases the formation of drops is accompanied by a proliferation of cells of the external enamel epithelium (Figs. 5 and 6). Both the adherent and the free drops located in the periodontium move upward with the growth of teeth. If, in the course of this movement, the ameloblasts, adjacent to the drops, remain intact, the drops become mineralized at the level of tooth where the enamel matrix normally undergoes maturation (Figs. 9, 12 and 13). On the other hand, when ameloblasts undergo distortion after the production of drops, the drops fail to calcify (Figs. 10, 12, 13 and 14). Sometimes in the same longitudinal section a repeated formation of drops can be observed. In such a case, on the same side of the tooth in the basal area of periodontium the drop is of organic matrix, while in the occlusal portion a calcified drop is to be seen. Both the uncalcified and calcified drops at a certain stage of their occlusal drift provoke the production of cementum masses at the level of tooth where periodontal tissue usually changes into columns of cartilage cementum. These masses show the structure of cartilage cementum (Fig. 12) or sometimes such osteocementum (Fig. 9).

All photomicrographs, with the exception of Fig. 2, were taken from preparations stained with hematoxylin and eosin and of the longitudinal section of lower molars or longitudinal and cross sections of lower incisors. In Figs. 2, 5, 13, and 18 the direction of tooth eruption is from the right to the left. In all other illustrations, with the exception of Fig. 10, the occlusal region is shown in the upper sector of the picture. All figures showing the formation of cementicles and of pseudocementicles relate to the functional part of tooth (tooth portion with completely formed enamel).

Fig. 1.—Vulnerability of junction of ameloblast layer and adjacent surface of growing enamel matrix "intravitam" and "post mortem." In area *x* is shown detachment of ameloblasts "intra vitam" followed by formation of enamel drops (dr). In lower portion of tooth detachment of ameloblast layer from enamel matrix occurred during histologic preparation. *Od.*, odontoblasts; *d.*, dentin; *E.*, enamel matrix; *Am.*, ameloblasts. Molar of guinea pig with subscurvy.

Fig. 2.—No "gap" evident between ameloblastic layer and surface of growing enamel matrix. Note zone of Tomes' processes. Hansen's stain. Molar of normal guinea pig.

Fig. 3.—Elevation and detachment of ameloblasts from enamel matrix with production of small adherent drops owing to overstress of incisor produced by forced opening of mouth by means of a wooden catch, through which catheter was introduced. Longitudinal section. Normal guinea pig.

Fig. 4.—Folding of ameloblast layer due to the trauma to incisor by repeatedly grinding off its crown. Longitudinal section. Normal rat.

Fig. 5.—Formation of free round enamel drop due to the trauma to incisor by repeatedly grinding off its crown. Detached ameloblasts have degenerated after the production of drop. Note proliferation of external enamel epithelium. Longitudinal section. Normal rat. 5*a*, Appearance of angle between scorbutic (x) portions of molor of guinea pig and those developed during the cure of scurvy (nr 5*a* is placed in this part). Border between this portion is shown by a coalescense of dentin with periodontium. Arrow indicates direction of eruption.

Fig. 6.—Development of adherent enamel drops due to the elevation of ameloblast layer from enamel matrix and production of free drops (marked with black lines) in periodontium from disrupted ameloblasts. Note proliferation of external enamel epithelium. Molar of guinea pig cured of scurvy.

Fig. 7.—Development of a free round large enamel drop due to folding of ameloblast layer. Note proliferation of external enamel epithelium. Molar of guinea pig cured of scurvy.

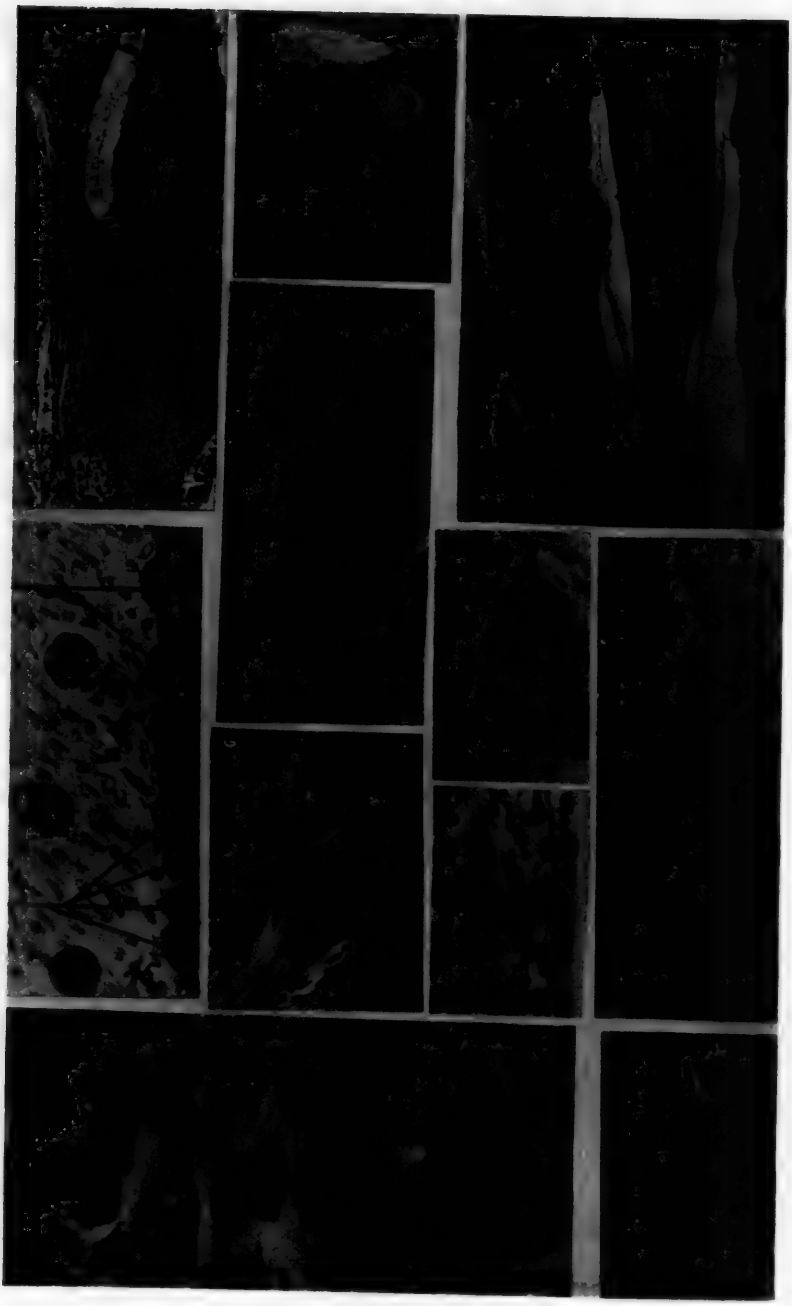
Fig. 8.—Detachment of ameloblast layer from enamel matrix caused by repeatedly grinding off an incisor's crown. Bleedings under the detached cell layer prove that this phenomenon is no artifact. Longitudinal section of the area where the enamel matrix has reached its ultimate thickness. Normal rat.

Fig. 9.—Mineralized free enamel drop on the level of tooth where normally the maturation of enamel matrix takes place. In the center of the drop there is interprismatic substance remaining after the decalcification during the making of the histologic preparation. Note the presence of ameloblasts at poles of drop and formation of osteocementum between drop and maturing enamel. Molar of guinea pig cured from scurvy.

Fig. 10.—Two uncalcified enamel drops on the level of tooth with mineralized enamel layer. The space between periodontium and dentin was formerly occupied by enamel now lost in decalcification. Note absence of ameloblasts at drops. Cross section of incisor of guinea pig with fluorosis.

Fig. 11.—Development of an adherent (in appearance like a fungus) and some free round (marked with black lines) enamel drops in a molar of a normal rabbit.

(For legends, see opposite page.)



(For legends, see opposite page.)

From these findings it may be concluded that the origin of enamel drops is determined by mechanic factors which provoke the strain or folding of the ameloblastic layer. On the other hand, the examination of teeth of guinea pigs with fluorosis shows that endogenous factors also may be significant in the formation of drops. In these animals the enamel drops sometimes develop in teeth in which foldings or the bending of dentin wall could not be observed with certainty. A further phenomenon of a multiple development of drops in the same jaw must be noted. Generally these drops do not calcify (Figs. 10 and 14). Since it has been assumed that fluorine directly affects the enamel-forming cells, it is very probable that in guinea pigs with fluorosis the instability of the junction of ameloblasts and growing enamel matrix is still more pronounced than in the teeth of normal animals.

The results of the present work show some details concerning the relation between the enamel and dentin during amelogenesis. Previous results of experiments of other workers[15, 44] and of the author[19, 22] have shown that the ameloblasts, in the early stage of their development, failed to produce enamel matrix, if they were separated from dentin[15] or if dentin was not formed at all.[19, 22, 44] On the other hand, the origin of free enamel drops shows that if the formation of the layer of enamel matrix had begun, the detached ameloblasts continued to produce enamel in periodontium (Figs. 1, 5, 7, 10, 12, 13 and 14). This phenomenon permits the conclusion that the presence of dentin is necessary only for the beginning of amelogenesis. In the later stage of enamel formation, the amelogenesis becomes independent of the presence of dentin.

Fig. 12.—Free large mature enamel drop. Note presence of the ameloblast layer covering the drop and the interprismatic substance in the center of the drop, which remained after the decalcification during the making of the histologic preparation. Under the drop and between this and the tooth wall many small enamel drops remained uncalcified. The development of drops caused the production of the large masses of cartilage cementum. Molar of guinea pig cured from scurvy.

Fig. 13.—Partly calcified enamel drop which became incorporated in the column of cartilage cementum in the course of its occlusal movement. The number of Fig. 13 is placed in space formerly occupied by enamel now lost in decalcification. Crown of molar of guinea pig cured from scurvy. Since the mature drops shown in Figs. 9, 12, and 13 are incorporated in the periodontium or cartilage cementum, they retained their shape almost unchanged also in the decalcified sections.

Fig. 14.—Multiple formation of enamel drops in fluorosis. All drops remained uncalcified on account of the distortion of the corresponding ameloblasts, and during their occlusal drift they became embedded in the column of cartilage cementum. Molar of guinea pig.

Fig. 15.—Free cementicles are located in periodontium and those interstitial are incorporated in primary cementum (last are indicated by black lines). Note nearly uniform size of cementicles. Molar of a normal rabbit.

Fig. 16.—Detachment of ameloblasts from growing enamel matrix and formation of a free enamel drop (over the number of the Fig.) caused the folding of badly calcified dentin in avitaminosis A. Longitudinal section of a rat's incisor.

Fig. 17.—Free and attached cementicles were located in the subepithelial connective tissue of gingiva owing to their occlusal movement in the fast erupting molar of a normal guinea pig.

Fig. 18.—The formation of a "pseudo-cementicle" in molar of guinea pig. Displaced cementum body, by means of which the fibers of periodontium are attached to the enamel in the functional portion of tooth. The number of Fig. 18 is placed in space formerly occupied by enamel now lost in decalcification.

Fig. 19.—The formation of a "pseudo-cementicle" due to displacement of a degenerated senile ameloblast into periodontium of functional part of tooth of guinea pig.

Fig. 20.—Formation of two large cementicles due to the conglomeration of many small ones. In upper part of illustration there is a large adherent cementicle and in lower part a large free one. Molar of a normal rabbit.

Fig. 21.—Lamellated structure of cementicle at magnification of about 1000×. Note inclusions of cells in center of cementicle and aggregation of some cementoblasts (on left side) which precedes development of cementicles. Molar of a normal rabbit.

Fig. 22.—Location of cementicle in epithelium of gingiva owing to its occlusal movement in continuously growing molar of a normal rabbit.

The findings show that cementicles develop only on the surface of tooth covered with cementum. Since in this area of the tooth of rodents the cells of enamel epithelium are never present the development of cementicles must be due to the action of cementoblasts only. The cementicles of rodents generally develop in the functional part of tooth and are shaped in the same manner as those of man. They are generally spherical of a concentric lamellated structure and sometimes show inclusions of cells (Figs. 15, 20, 21 and 22). Unlike enamel drops cementicles do not cause the production of masses of cartilage cementum or osteocementum. Since no considerable difference was found between the origin of cementicles in normal animals or in those with avitaminosis A or C, or with hormonal disturbances, and since they were found particularly in animals fed with a catheter it may be assumed that in rodents, as in man, the origin of cementicles is due to overstress of the periodontal tissue. This postulation may be supported also by the fact that no cementicles could be found in the formative portion of rodent teeth. Overstress causes a more or less pronounced dislocation of cementoblasts into the periodontium. If this displacement is slight, adherent cementicles are developed (Figs. 15 and 20) and during the growth of the primary cementum in width, they are embedded in the primary cementum and form part of the newly deposited cementum layers. These are the so-called "interstitial" cementicles (Fig. 15). From cementoblasts which are more markedly dislocated in the periodontium free cementicles originate (Figs. 15, 17, 21 and 22). Usually the cementicles develop to an almost uniform size. If simultaneously, several groups of cementoblasts are detached, large cementicles may be produced through the conglomeration of small ones (Fig. 20). When cementoblasts aggregate cementicles with inclusions of more or less cells are formed (Fig. 21). Owing to the upward motion with the eruption of teeth, the cementicles locate in the epithelium of gingiva (Figs. 17 and 22). Cementicles are more often found in rabbits than in guinea pigs or rats.

In the region of guinea pig periodontium adjacent to the tooth portion covered with enamel, cementum bodies which have no connection with the formation of enamel drops or cementicles sometimes appear. Usually these bodies are found on the enamel and serve as points of anchorage by means of which the fibers of the periodontium are attached to the surface of mature enamel. Due to overstress, these bodies disrupt from the enamel and dislocate into the periodontium (Fig. 18). Occasionally single degenerated senile ameloblasts are displaced into the periodontium and, like enamel drops, provoke calcification of the surrounding tissue, which has the appearance of small cementum bodies (Fig. 19). Since genuine cementicles are produced by cementoblasts, these atypical cementum bodies may be called pseudocementicles.

SUMMARY AND CONCLUSIONS

The origin of enamel drops and cementicles was studied in the continuosly growing teeth of guinea pigs, rabbits and rats.

1. Enamel drops are formed by active ameloblasts, detached from the surface of growing enamel matrix.

2. Genuine cementicles originate from cementoblasts, dislocated from primary cementum into the periodontium.

3. In the region of the periodontium, adjacent to that portion of the tooth covered with enamel, atypical cementum bodies (pseudo-cementicles) which have no connection with the origin of enamel drops and cementicles sometimes appear.

4. The origin of free enamel drops shows that the presence of dentin is necessary only for the beginning of amelogenesis.

The chief part of the experiments was performed at the Department of Pharmacology, University of Upsala, Sweden, to the former head of which, Professor E. L. Backman, the author wishes to express his sincerest appreciation for the help granted and the interest shown in this work.

REFERENCES

1. Agnew, G.: Microscopic Studies on Paradental Tissue, *J. D. Res.* **12**: 442, 1932.
2. Bauer, W.: Mechanischerzeugte Entwicklungsstörungen des Zahnkeimes (''Experimentelle Zahnkeimmissbildungen''), *Deutsche Mtschrft f. Zhlkde* **46**: 737, 1928.
3. Bauer, W.: Ueber Zementikel und zementikelähnliche Einlagerungen in der Wurzelhaut, *Vierteljschr. Zhlkde* **45**: 345, 1929.
4. Baume, R.: *Lehrbuch der Zahnheilkunde* 3. (Aufl. Leipzig 1890)
5. Bernier, J. L., and Thompson, H. C.: The Histogenesis of the Cementoma., *Am. J. Orthodont. and Oral. Surg.* **32**: 543, 1946.
6. Boyle, P. E.: Tooth Suspension. A Comparative Study of the Paradental Tissues of Man and of the Guinea Pig, *J. D. Res.* **17**: 37, 1938.
7. Böhm, G.: Schmelztropfen mit besonderer Berücksichtigung des histologischen Baues, *Deutsche Zahnärztl. Wschrft.* **41**: 1013, 1938.
8. Dewey, K. W.: Normal and Pathological Cementum, *D. Cosmos* **68**: 560, 1927.
9. Euler-Meyer: Pathohistologie der Zähne mit besonderer Berücksichtigung der Pathobiologie (München, 1927).
10. Gottlieb, B.: Zur Ätiologie und Therapie der Alveolar-Pyorrhoe, *Öst. Ztchr. Stomatol.* **46**: 215, 1920.
11. Gottlieb, B.: Zementexostosen, Schmelztropfen und Epithelnester, *Ztschr. Stomatol.* **19**: 40, 1921.
12. Goldman, H. M., and Bloom, J.: A Collective Review and Atlas of Dental Anomalies and Diseases, *Oral. Surg., Oral Med. & Oral Path.* **2**: 874, 1949.
13. Göllner, L.: Ueber Schmelztropfen, *Deutsche Mtschr. Zhnhlke* **46**: 225, 1928.
14. Höpfel, W.: Ueber Zementikelbildungen, *Deutsche Zahnärztl. Wschr.* **39**: 741, 1936.
15. Huggins, C. B., Carrol, H. B., and Dahlberg, A. A.: Heterotopic Formation of Dentine and Enamel by Transplantation of Epithelium and Pulp Tissue, *Proc. Soc. Exper. Biol. & Med.* **31**: 525, 1934.
16. Jonson, G., Obel, A. L., and Sjöberg, K.: Skorbut als Sekundärerscheinung bei A-vitaminose I, *Ztschr. Vitaminf.* **12**: 300, 1942.
17. Jonson, G., Obel, A. L., and Sjöberg, K.: Skorbut als Sekundärerscheinung bei A-vitaminose II, *Ztschr. Vitaminf.* **15**: 115, 1944.
18. Kantorowicz, F. A.: Ueber den Bau und die Entstehung der Schmelztropfen, *Deutsche Mschrft. Zhnhlkde* **22**: 17, 1904.
19. Kalnins, V.: Wirkung der Ascorbinsäure auf die Zähne und den Kieferknochen skorbutischer Meerschweinchen. *Upsala Läkaref. förh.* **45**: 117, 1939.
20. Kalnins, V.: Experimente über die Wirkung der Organpräparate Vaduril und Alveoaktiv auf die Zähne und das Paradentium, *Upsala Läkaref. förh.* **52**: 369, 1946.
21. Kalnins, V.: Ueber den Vitamin C-Bedarf des Kaninchens. *Upsala Läkaref. förh.* **51**: 215, 1946.
22. Kalnins, V.: Developmental Disturbances of Enamel in Scurvy, *J. D. Res.* **31**: 440, 1952.
23. Kalnins, V., und Ledina, H.: Die Wirkung des Epithelkörperchenshormons auf Zähne und Knochen normaler und skorbutischer Meerschweinchen. *Upsala Läkaref. förh.* **52**: 235, 1947.
24. Kreshaw, J.: An Enamel Nodule, *Brit. D. J.* **2**: 75, 1934.
25. Kronfeld, R.: The Biology of Cementum, *J. A. D. A.* **25**: 1451, 1938.
26. Lartschneider, J.: Beiträge zur Kenntnis der für die Entstehung und die Lokalisation der ''Schmelztropfen'' massgebenden biologischen und pathologischen Momente, *Ztschr. Stomatol.* **27**: 751, 1929.
27. Loos, R.: Cementhyperplasie; Schemlztropfen; äussere Odontome, *Scheffs Handbuch der Zahnheilkunde* **1**: 600, 1902.
28. Malassez, M. L.: Sur le role des débris epithéliaux paradentaires, *Arch. de Physiol.* **5**: 129, 1885; **6**: 378, 1885.

29. Malassez and Gallipe: Cited by Willinger: Notes sur "les perles d'email" (Stomatologie Nr 8, 1908). *Deutsche Mtschr. Zhnhlkde,* 1910.
30. Micola, O. J., and Bauer, W. M. K.: "Cementicles" and Fragments of Cementum in the Periodontal Membrane, *Oral Surg., Oral Med. & Oral Path.* **2**: 1063, 1949.
31. Münch, J.: Pathologische Histologie der Zähne, *Fortschritte der Zhnhlkde* **5**: 259, 1929.
32. Öhnell, H., Westin, G., and Hjärre, A.: Studie über Skorbut und Fluorose bei Meerschweinchen, *Acta Pathol. Microbiol. Scand.* **13**: 1, 1936.
33. Orban, B.: Ein Besonderer Befund an einem Schmelztropfen, *Ztschr. Stomatol.* **22**: 188, 1924.
34. Palazzi, S.: Die harten Neubildungen des Paradentiums, *Zahnärztl. Rundsch.* **48**: 155, 1939/2.
35. Pflüger, H.: Entstehung der Schmelzperlen, *Deutsche Mtschr. Zhnhlkde* **49**: 993, 1931.
36. Ponte, J. J. R.: Perlas adamantinos (Buenos Aires) *Rev. odontolog* **8**: 469, 1928.
37. Rywkind, A. W.: Zur Frage der Zementikelbildung *Ztschr. Stomatol.* **28**: 1178, 1930.
38. Schmamine, F.: Das Sekundäre Zement, *Deutsche Zahnheilkunde* (13., Heft., 1910). Cited by Bauer (Reference, No. 3).
39. Schlenker, M.: Schmelztropfen, äussere Odontome usw. *Scheffs Handbuch der Zahnheilkunde* **1**: 529, 1891.
40. Thoma, K.: *Oral Pathology* (Cementicles p. 431 and Enameloma p. 946), St. Louis, 1944, The C. V. Mosby Co.
41. Thoma, K., Cementoblastoma, *J. Orthodontia* **51**: 1127, 1946.
42. Thoma, K., and Goldman, H. M.: The Pathology of Dental Cementum, *J. A. D. A.* **26**: 1943, 1939.
43. Walkhoff, O.: Bau und Entstehung eininger Zahnmissbildungen, *Deutsch. Mtschr. Zhnhlkde* **14**: 261, 1896.
44. Wasserman, F.: Analysis of the Enamel Formation in the Continuously Growing Teeth of Normal and Vitamin C Deficient Guinea Pigs, *J. D. Res.* **23**: 463, 1944.
45. Weski, O., und Contrares, D.: Befunde und Vorgänge im periradikulären Raume der mehrwurzeligen Zähnen, *Vrtljschr. Zhnhlkde* **40**: 229, 1924.
46. Wedl, C.: Über Gefässknäuel im Zahnperiost, *Virch. Arch.* **85**: 175, 1881.
47. Weinmann, J. P.: Recovery of ameloblasts, *J. A. D. A.* **30**: 874, 1943.

SALIVARY BACTERIA. III. THE PATHOGENICITY OF ORAL STAPHYLOCOCCI

ARNOLD J. SINGER, PH.D.

Jersey City, N. J.

THE gram-positive micrococci are among the common components of the oral flora. This family of bacteria is extremely diverse and includes the loosely defined group of staphylococci. The pathogenicity of the staphylococci has never been defined and whether there are saprophytic and parasitic forms, or whether the saprophytes are potential pathogens has defied clarification. There have been many attempts to classify the staphylococci in order to distinguish between those of the group which are pathogens or potential pathogens and those of a saprophytic nature. The earliest classification was based on the chromogenicity of these cocci and they were identified as pigmented or non-pigmented organisms. However, variations in pigment production were so great that recognition had to be given to degrees of pigmentation. It has been clearly shown, however, that pathogenicity and chromogenicity are not correlatable phenomena,[6] even though the yellow pigment producers are frequently associated with pathologic processes.

Another characteristic of these organisms which has been exhaustively studied is their fermentation reactions. The staphylococci cannot be specifically identified by their behaviour toward certain carbohydrates. These organisms vary considerably in their fermentation reactions and, on culture, seem to change their biochemical characteristics in an unpredictable way. However, it has been reported that many of the pathogenic staphylococci ferment mannitol and this characteristic has been used as an aid in classifying these organisms.[3]

Some correlation between gelatin liquefaction and pathogenicity has been reported for the staphylococci.[18] The ability of some staphylococci to coagulate oxalated plasma[6, 11, 15] has been studied in considerable detail. This property which has been ascribed to an enzyme, coagulase, has also been found to be correlated to some extent, at least, with pathogenicity.[5, 7] Since the staphylococci are extremely resistant organisms attempts have been made to correlate pathogenicity with their ability to grow on special media containing dye inhibitors but no real correlation has been found to exist.

Finally, the toxigenic properties of these bacteria have also been widely studied[2, 12] since the potency and diversity of these toxins have indicated that some relationship between toxicogenesis and pathogenicity probably exists. Some staphylococci produce toxins which are capable of hemolyzing red blood cells of many animal species and man (hemolysin); of causing extensive dermal necrosis after intradermal injection of minute quantities (necrotoxin or dermal necrotizing toxin); of rapidly killing rabbits by respiratory failure after intravenous injection of minute amounts (lethal toxin); of destroying white blood cells (leukocidin); and of prolonged stimulation of the vomiting centers of the brain causing vomiting in those animals subject to this reflex excitation (entero-

Received for publication, January 5, 1952; revised by author, May 13, 1952.

toxin or food poisoning toxin). The early voluminous literature which has accumulated on these various phenomena has been adequately reviewed by Blair.[1] While these organisms have received considerable attention in the general and medical bacteriologic literature, they have not attracted the attention of many of those interested in oral bacteriology. The literature reviews of Hadley[10] and Graber[9] indicate many references to staphylococci but the pathogenicity or biochemistry of the identified organisms has not been investigated.

These organisms have many properties that are intriguing with regard to oral pathology. Maximum toxigenesis is obtained in filtrates when the organisms are grown in an environment containing an increased carbon dioxide tension and such an environment is supplied in saliva. The organisms are also capable of coagulating protein and are characteristically focal point invaders. They are one of the organisms most frequently encountered in osteomyelitic conditions. Their acidogenic power is equal to that of other microorganisms commonly found in the mouth.[8]

This investigation was then undertaken to determine the incidence of staphylococci among the gram-positive micrococci of the saliva and to determine the occurrence of those biochemical properties ordinarily associated with pathogenicity of staphylococci.

PROCEDURES

Unstimulated saliva from unselected individuals was used for the isolation of the organisms. The saliva was collected in sterile shell vials and 0.1 ml. was inoculated by streaking onto the special media for primary isolation.

Staphylococcus medium No. 110 (Difco) was used for primary isolation. This medium is selective for staphylococci and also permits for the identification of several characteristics of the staphylococci. In these studies only gram-positive micrococci, morphologically identical with staphylococci, were isolated on this medium.

The medium for toxin production was the semisolid agar of Leonard and Holm.[14] This medium was sterilized, distributed into sterile Petri dishes in 20 ml. quantities and inoculated from a broth subculture from the primary isolation. For toxin production, the Petri dishes were placed in a desiccator with a vacuum attachment, the air was evacuated and replaced with a mixture of 30 per cent CO_2 and 70 per cent air, and the desiccator valve was then closed to maintain the environment. After 48 hours of incubation at 30° C., the Petri dishes were removed and the agar separated by centrifugation. The clear supernatant was decanted and used as the exotoxin containing filtrate.

Pigment production was determined by placing a loopful of 24 hour culture from an agar slant onto a white porcelain spot plate and the organisms classified as white or yellow.

Mannitol fermentation was identified on the plates of primary isolation by dropping bromocresol purple indicator on representative colonies. A color change of the indicator was indicative of mannitol fermentation.

Gelatin liquefaction was also determined on the medium of primary isolation by flooding the plates with a saturated ammonium sulfate solution. Gelatin

liquefaction was indicated by clear or plaque zones in the medium around the colony.

Since hemolysin titer was not of primary interest, the ability of the organisms to hemolyze human red blood cells was measured on blood-agar plates rather than by titration. In each case the alpha hemolysin property was determined. No effort was made to measure beta hemolysin. The blood agar plates were pour plates inoculated from a 24 hour broth subculture from the primary isolation.

Ureolysis of the isolated organisms was also investigated because of the reports on the potential value of urea in the oral environment. The method for screening ureolytic organisms has been previously described.[17]

The dermal necrotizing effect of the filtrates was measured by injecting 0.2 ml. of centrifuged filtrate intradermally into the abdominal wall of guinea pigs or rabbits which had been previously epilated. Necrotoxin containing filtrates produced a very characteristic response. Within several hours after injection, the filtrate diffused through the skin and the area assumed a bluish, bruised appearance with a red marginal area. In 24 to 48 hours, the skin became necrotic and sloughing ensued. In 7 to 10 days, the necrosis followed the typical pattern of a necrotizing infection with complete healing and scar tissue formation. Using guinea pigs, as many as four filtrates can be tested simultaneously by quadranting the epilated abdomen. With rabbits, eight filtrates can be tested at one time, four on either side of the abdomen. In either case, occasional merging of lesions may occur with extremely potent filtrates. Since these toxins are antigenic, it is probable that the animals build a slow immunity to the filtrates. They have been found satisfactory, however, for several repeated injections.

EXPERIMENTAL

In this study of the incidence and nature of the oral staphylococci, the saliva of 53 individuals was plated on the staphylococcus medium. The plates were incubated for 24 hours and typical colonies selected for microscopic study by gram stain. Subcultures were then made for further study and the inoculum for the coagulase test removed. The other tests outlined in the preceding section were performed.

The results of the studies on all the staphylococci isolated are presented in Table I. Only one of the 53 individual salivas failed to yield staphylococci on primary isolation. From some individuals, colonies of different morphology were seen on primary culture. Each variation was subcultured giving a total of 89 isolations for study.

Of the 89 organisms, 41 were pigmented and 48 nonpigmented, 10 fermented mannitol, 66 liquefied gelatin, 16 produced the enzyme coagulase, 45 produced hemolysin, 31 were dermonecrotoxicogenic, and 40 produced urease to split urea. Twelve of the 16 coagulase producers also produced necrotoxin and four of the mannitol fermenters were also coagulase and necrotoxin positive. It is obvious therefore that no definite pattern of biochemical properties of oral staphylococci could be demonstrated.

TABLE I. THE BIOCHEMICAL AND TOXIGENIC PROPERTIES OF STAPHYLOCOCCI ISOLATED FROM SALIVA

ORGANISM ISOLATED FROM SUBJECT	PIGMENT	MANNITOL FERMENTATION	GELATIN LIQUEFACTION	TOXIGENICITY			
				COAGULASE	HEMOLYSIN	NECROTOXIN	UREOLYSIS
1	yellow	–	+	–	–	–	4+
1A	white	–	+	–	+	+	4+–
2	yellow	–	+	–	–	–	–
3	yellow	–	2+	–	+	2+	4+
4	yellow	–	2+	4+	–	2+	–
4A	white	–	2+	–	–	+	–
5	yellow	–	2+	4+	–	4+	–
5A	white	–	2+	–	–	+	4+
6	white	–	+	–	–	2+	4+
7	white	–	+	–	+	–	4+
8	yellow	–	2+	–	–	–	–
8A	white	–	2+	–	–	–	2+
9	white	–	+	–	–	–	2+
10	yellow	–	+	4+	–	–	–
11	cream	–	+	–	–	2+	–
12	white	–	+	–	–	–	4+
13	yellow	–	–	–	+	+	4+
14	white	–	+	–	+	–	4+
14A	orange	+	+	4+	+	+	–
14B	cream	+	+	–	–	–	–
15	white	–	+	–	–	+	4+
16	white	–	+	–	–		4+
17	yellow	–	+	4+	+	4+	–
17A	cream	–	+	–	–	–	4+
18	cream	–	+	–	–	–	4+
18A	white	–	–	–	+	–	4+
19	white	–	+	–	0	–	–
20	orange	–	+	4+	–	–	4+
20A	white	–	+	–	+	–	4+
21	white	–	+	–	+	–	4+
22	white	–	+	–	–	–	4+
22A	yellow	–	+	4+	+	2+	–
23	yellow	+	4+	–	–	–	2+
23A	white	+	–	–	–	–	4+
24	yellow	+	+	4+	+	4+	–
24A	white	–	+	–	+	–	4+
25	white	–	+	–	+	+	4+
26	white	–	–	–	+	–	2+
26A	yellow	–	+	–	+	–	–
27	yellow	+	+	–	–	–	–
27A	white	–	+	–	+	2+	2+
28	white	–	+	–	–	–	–
29	yellow	–	–	4+	0	2+	–
29A	white	–	–	–	–	–	–
30	cream	–	–	–	0	–	–
31	white	–	+	–	–	–	2+
32	No growth on primary isolation						
33	white	–	+	–	–	+	4+
33A	cream	–	–	4+	–	+	–
34	white	–	+	–	+	+	–
34A	cream	–	+	–	–	–	3+
35	yellow	–	+	–	–	+	–
35A	white	–	+	4+	+	–	4+
35B	cream	–	+	–	+	–	–
36	yellow	–	+	–	–	–	+
36A	cream	–	–	–	–	–	–

TABLE I—CONT'D

ORGANISM ISOLATED FROM SUBJECT	PIGMENT	MANNITOL FERMENTATION	GELATIN LIQUEFACTION	TOXIGENICITY			
				COAGULASE	HEMOLYSIN	NECROTOXIN	UREOLYSIS
36B	white	–	–	–	–	–	–
37	yellow	–	+	–	–	–	–
37A	white	–	+	–	2+	–	–
38	cream	–	+	–	+	–	–
38A	white	–	+	–	2+	–	–
39	white	–	–	–	+	–	–
40	white	+	+	–	+	–	–
40A	yellow	–	+	4+	+	–	2+
41	yellow	–	+	–	–	–	–
41A	white	–	+	–	2+	–	–
41B	white	–	+	–	+	–	+
42	cream	–	+	–	+	–	–
42A	cream	–	+	–	2+	–	+
43	yellow	–	–	4+	+	2+	2+
43A	white	–	+	–	+	–	–
43B	white	–	+	–	–	–	–
44	white	–	+	–	+	–	–
44A	white	–	+	–	–	2+	–
45	cream	–	+	–	+	–	3+
45A	cream	–	–	4+	4+	3+	+
46	yellow	–	+	–	–	+	–
46A	white	+	–	–	–	+	4+
47	cream	+	+	4+	+	+	–
47A	white	–	+	–	3+	–	–
48	yellow	–	–	–	+	–	–
48A	white	–	–	–	+	–	–
49	white	+	+	4+	2+	4+	–
50	white	–	–	–	+	+	–
50A	white	–	–	–	2+	–	–
51	white	–	–	–	+	–	–
52	yellow	–	–	–	–	–	–
52A	white	–	–	–	–	+	–
53	white	–	–	–	2+	+	2+
53A	white	–	–	–	3+	–	2+

It is also interesting to note that if Stone's test for gelatin liquefaction is an accurate test for pathogenicity, then 66 of the isolated staphylococci can be classed as pathogens. On the other hand, if mannitol fermentation is taken as a criterion of pathogenicity, then only 10 of the 89 organisms isolated are pathogenic and only 16 if coagulase production is used for classification. If all of the proposed tests are used as a cumulative criterion then only 4 organisms would be considered pathogenic.

EFFECT OF UREA ON TOXIGENESIS

Nine representative organisms were selected from each of the following groups: ureolytic and necrotoxigenic, ureolytic but not necrotoxigenic, and necrotoxigenic but not ureolytic. Studies were then made on the filtrates for toxic properties and the results are presented in Table II.

The results indicated that ureolytic, toxigenic organisms produced potently toxic filtrates in the absence of urea. But when urea was added to the

media, either toxin production was impaired or the toxic metabolites were hydrolyzed due to the liberated ammonia. On the other hand, urea did not induce toxigenesis in organisms which normally were not toxigenic, nor did it affect toxin production in nonurolytic organisms.

DISCUSSION

While there have been a number of reports in the literature on the presence of staphylococci in the oral environment, there has been no demonstration of the potential pathogenicity of these normal oral inhabitants. This investigation has shown that in a group of 53 adults only one individual did not yield staphylococci at the time of survey. It does seem as though the properties of the staphylococci and their apparent widespread incidence are worthy of further investigation.

The incidence of these toxigenic organisms in those oral infections accompanied by necrosis would be of interest since these toxic filtrates could very easily contribute to oral disease. In dental caries, these organisms may also play a contributory role. They form localized infections, are proteolytic and produce acid from carbohydrate. This is of particular interest in view of the recent reports by Manly.[16] He has shown, histologically, that an area of acid decalcification may be followed by bacterial invasion along the pathways of the lamellae. The subsequent cariogenic process, it is suggested, may be due to proteolytic activity by the invading organisms.

Acidogenic, proteolytic staphylococci are normally present in the oral environment. These organisms show the same variations exhibited by staphylococci isolated from many different sources.

Of the 89 organisms isolated 31 were producers of toxic filtrates capable of producing a dermal necrosis following intradermal injection. This necrotizing effect is similar to the necrosis observed with some acute infections and follows

TABLE II. EFFECTS OF THE ADDITION OF UREA TO THE MEDIUM ON THE TOXICOGENESIS OF UREALYTIC AND NONUREALYTIC ORGANISMS

ORGANISM	UREASE+ NECROTOXIN+	ORGANISM NO.	UREASE+ NECROTOXIN−	ORGANISM NO.	UREASE− NECROTOXIN+
1A	Not Necrotoxic	1	Not Necrotoxic	4	2+ Necrotoxin
3	Not Necrotoxic	7	Not Necrotoxic	5	2+ Necrotoxin
5A	Not Necrotoxic	8A	Not Necrotoxic	14A	1+
6	Not Necrotoxic	9	Not Necrotoxic	17	2+
13	Not Necrotoxic	12	Not Necrotoxic	22A	2+
15	Not Necrotoxic	23	Not Necrotoxic	24	1+
25	Not Necrotoxic	24A	Not Necrotoxic	29	+
27A	Not Necrotoxic	26	Not Necrotoxic	33A	2+
33	Not Necrotoxic	28	Not Necrotoxic	35	2+
		31	Not Necrotoxic	47	2+
				49	1+

the same pattern with sloughing and scar tissue formation. This necrotizing toxin, and the other toxins produced by the staphylococci, are produced only in the presence of an increased CO_2 tension. It has been suggested that the role of the CO_2 is that of buffer and the similarity between the oral environment with the increased CO_2 tension and the CO_2 requirement for toxin production by the staphylococci lends for interesting speculation.

CONCLUSIONS

Among the gram-positive micrococci of the oral environment, the staphylococci are commonly present. These organisms conform in all respects to the criteria for the staphylococci including the diversity of biochemical properties. There has been no definite pattern of biochemical properties observed. The oral staphylococci show the same vagaries as staphylococci reported from other sources.

It has also been observed that either urea inhibits toxigenicity of ureolytic, toxigenic organisms or the alkalinity resulting from ureolysis hydrolyzes the toxic metabolites. In either case, the addition of urea to the environment renders the filtrates of these staphylococci innocuous.

REFERENCES

1. Blair, J. E.: The Pathogenic Staphylococci, *Bact. Rev.* **3**: 97, 1939.
2. Burnet, F. M.: The Exotoxins of Staphylococcus Pyogenes aureus, *J. Path. Bact.* **32**: 717, 1929.
3. Chapman, G. H.: The Significance of Sodium Chloride in Studies of Staphylococci, *J. Bact.* **50**: 201, 1945.
4. Chapman, G. H.: A Single Culture Medium for Selective Isolation of Plasma-coagulating Staphylococci, *J. Bact.* **51**: 409, 1946.
5. Chapman, G. H., Berens, C., Nilson, E. L., and Curcio, L. G.: The Differentiation of Pathogenic Staphylococci from Non-pathogenic Types, *J. Bact.* **35**: 311, 1938.
6. Chapman, G. H., Berens, C., Peters, A., and Curcio, L.: Coagulase and Hemolysin Tests as measures of the Pathogenicity of Staphylococcus, *J. Bact.* **28**: 343, 1934.
7. Cruickshank, R.: Staphylocoagulase, *J. Path. & Bact.* **45**: 295, 1937.
8. Florestano, H. J.: Acidogenic Properties of Certain Oral Microorganisms. *J. D. Res.* **21**: 263, 1942.
9. Graber, T. M.: Local Factors in Dental Caries, *J. A. D. A.* **35**: 92, 1947.
10. Hadley, P.: The Bacteriology of Dental Caries, *D. Cosmos* **66**: 707, 1924.
11. Hallman, F. A.: Pathogenic Staphylococci in the Anterior Nares: Their Incidence and Differentiation, *Proc. Soc. Exper. Biol. & Med.* **36**: 789, 1937.
12. Kellaway, C. H., Burnet, F. M., and Williams, F. E.: The Pharmacological Action of the Exotoxin of Staphylococcus aureus, *J. Path. & Bact.* **33**: 889, 1930.
13. Lefkowitz, Wm., and Singer, A. J.: The Effect of Urea and Diammonium Phosphate on the pH of the Tooth Surface, *N. Y. State D. J.* **17**: 159, 1951.
14. Leonard, G. F., and Holm, A.: A Method for the Production of Staphylococcus Toxin and Toxoid, *J. Immunol.* **29**: 209, 1935.
15. Loeb, L.: The Influence of Certain Bacteria on the Coagulation of the Blood, *J. Med. Res.* **10**: 407, 1903.
16. Manly, E. B.: Caries of the Enamel. I. The Significance of Enamel Lamellae, *Brit. D. J.* **91**: 36, 1951.
17. Singer, A. J.: Salivary Bacteria. II. The Ureolytic Activity of Micrococci Isolated From Saliva, *Oral Surg., Oral Med., & Oral Path.* **4**: 1568, 1951.
18. Stone, R. V.: A Cultural Method for Classifying Staphylococci as of the "Food Poisoning Type," *Proc. Soc. Exper. Biol. & Med.* **33**: 185, 1935.

Erratum

In the June issue of the JOURNAL, Part X of the Evanston Dental Caries Study (p. 346), by I. N. Hill, J. R. Blayney, and Walter Wolf, footnotes to Tables IV to VIII erroneously included the word "not." The footnotes should read, "Differences are statistically significant."

INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

Scientific Proceedings of the Pacific Northwest Section

Spring Meeting, April 19 to 20, 1952

B. O. A. Thomas, *Editor, Pacific Northwest Section*

On Saturday evening, a round-table discussion was held, the topic being "Synthetic Resinous Filling Materials." The Program was as follows: Dr. John I. Ingle, Pulp Reaction to Synthetic Resins; Mr. Herbert Gaskill, Chemical and Physical Properties of Synthetic Resins; Drs. Ellis Jump and A. G. Maudsley, Discussion.

The regular Scientific Section was held at the University of Washington School of Dentistry on Sunday, April 20. Abstracts from this meeting follow.

1. Tooth Changes in a Case of Osteopetrosis. *B. O. A. Thomas, David B. Law, and Robert Johnson.* Five primary teeth were extracted from a 14-month-old female child suffering from osteopetrosis. Skeletal and oral radiographs showed bone changes diagnostic of this disease. Laboratory studies confirmed the radiographic diagnosis. The child presented with marked gingivitis and caries of several teeth. Ground sections were prepared from 2 teeth and decalcified sections from the remaining 3 teeth. The only significant alteration from normal was a metaplasia of the pulp tissue in 2 of the teeth. In these specimens postnatal dentin formation was very irregular, especially at the apical regions. Calcification of fibrous connective tissue along the pulpal surface of the dentin resulted in irregular "spurs" of nontubular tissue. It is possible that this metaplasia was a reaction to the dental caries, rather than a change associated with osteopetrosis. An attempt is being made to secure further information on this case. The author was unable to find anything in the literature pertaining to tooth changes in human osteopetrosis.

2. Discrepancies in Tooth Size. *M. Wayne Bolton.* Fifty-five cases of excellent occlusions were examined. Certain measurements of tooth widths were recorded; relations of overbite, overjet, and cusp height and incisor lengths were compared and correlations were calculated. Conclusions regarding these various factors were summarized and an anterior ration and a posterior relation for tooth size was developed. The necessity for a diagnostic setup in cases of doubt comparing the possible relation and interdigitation of maxillary and mandibular teeth was pointed out.

3. Histological Findings in the Kidney, Liver, Pancreas, Adrenal and Thyroid Gland of the Rat Following Sodium Fluoride Administration. *Alfred L. Ogilvie.* An experiment was conducted involving the intraperitoneal administration of sodium fluoride to 20 rats and the subsequent study of the pancreas, kidney, liver, adrenal gland, and thyroid gland of each of these animals. At autopsy no significant changes were observed either in the gross appearance of the animals (incisor teeth excepted) or in the gross structure of their viscera. Definite modifications were observed, however, in the microscopic structure of the kidney, pancreas, and thyroid gland. In the kidney, an edema was produced within the connective tissue between the tubules of the lower half of the papilla. Both the glomeruli and the medulla gave evidence of hyperemia and a possible increased vascularity. The pancreatic acinar cells showed an increased mitotic division. The interlobular and intralobular septa of the pancreas in treated animals were of greater width. The thyroid gland exhibited a mild atrophy of follicle epithelium involving a decrease in the size of the follicle cell nuclei and the cellular width. However, the height of the columnar epithelial cells of the follicle remained unaltered. There was evidence of an increase in adrenal gland weight and of adrenal cortical hypertrophy. The liver was not histologically changed.

4. Clinical and Histological Reactions of the Healthy Dental Pulp to Paraffin and to a Polyantibiotic Material Used During Pulpotomy. *Bill Seims.* The purposes of this study were, first, to ascertain the ability of healthy dental pulp tissue to heal under a bland, nonirritating dressing, and secondly to study the pulp's reaction to a polyantibiotic material preliminary to the latter's employment in contaminated cases. Fifty vital pulp amputations were performed on healthy, vital permanent first premolars and deciduous canines. Patients ranged from 7 to 14 years of age, each with 2 or more teeth treated. The period of observation ranged from 2 to 18 weeks. Paraffin and the concentrated polyantibiotic PBSC were selected for the capping materials. Analysis of the clinical and radiographical findings following pulpotomy indicated a successful result from using paraffin and PBSC as capping agents insofar as eighteen weeks of observation could determine. No teeth showed evidence of periodontal involvement, postoperative disturbances, or malfunction of the teeth. In half of the cases the pulps were examined histologically. They showed a fibrous tissue or osteoid barrier forming across the exposed area, but due to the presence of inflammation, degeneration or metaplasia, a hopeful prognosis seemed doubtful. The use of PBSC and paraffin has not been demonstrated to be of greater value than calcium hydroxide or several other agents.

5. Stomatosis Plicata. *F. G. Everett and H. J. Noyes.* A most unusual oral lesion is reported in two brothers, young white adults. The mucous membrane of the cheeks, the vestibular surface of the lips, the fornices of the vestibulum of the mouth, and the under surface of the tongue exhibit a soft whitish, folded appearance, and look as if parboiled. There is occasional desquamation of the epithelium. The lesions are painless except on consumption of spicy or acid foods. The condition was first noted in both brothers at about 13 years of age. Clinical and laboratory findings were largely negative, as were the results of antibiotic and antihistaminic therapy. Histologic examination showed the major changes to be present in the epithelium with a gross disturbance of the normal maturation pattern. With exception of the stratum germinativum which was fairly normal, all other epithelial layers showed pyknosis of nuclei, hydropic degeneration and vacuolization, occasional absence of nuclei, and bizarre cell forms. There was a wide scattering of extranuclear basophilic bodies in the cytoplasm of the stratum spinosum and outward, taking special keratohyalin stain. Hydropic degeneration had led to the formation of small intra-epithelial vesicles in places. The basement membrane was intact. The lamina propria showed infiltration with round cells and polymorphonuclear neutrophile leukocytes. There was also found a considerable number of mast cells and a few polymorphonuclear basophile leucocytes. The connective fiber bundles in the deeper layers of the propria appeared homogenized and broken up in places. In the opinion of the authors the lesions are of a chronic degenerative and possibly congenital nature. The name of stomatosis plicata for this condition is suggested.

INTERNATIONAL ASSOCIATION FOR DENTAL RESEARCH

Scientific Proceedings of the Minnesota Section

Spring Meeting, April 18, 1952

S. A. Lovestedt, *Secretary, Minnesota Section*

1. Multiple Myeloma Occurring in the Jaws: A Study of 17 Cases, *Karl W. Bruce* and *R. Quentin Royer, Mayo Clinic, Rochester, Minn.* The case records and the roentgenograms of the jaws of 59 patients with multiple myeloma were reviewed. Seventeen of these patients exhibited evidence in the

jaws. Among these 59 patients the ratio of males to females was approximately 2 to 1. However, among the patients with lesions in the jaws more females than males were affected. In all but one case of myeloma of the jaws there was general skeletal involvement. One case represented solitary myeloma of the jaws, and there was no evidence of lesions elsewhere in the body. The mandible was more frequently involved than the maxilla. Lesions occurred more often in the posterior portion of the body, the angle, and the ascending ramus of the mandible than elsewhere in the jaws. More patients exhibited lesions in the skull than in the jaws. Symptoms referable to the jaw, such as pain, pressure sensation, numbness, swelling, expansion of the jaw, mobility of teeth and epulis formation, were observed. In 7 cases the initial symptoms of myeloma were observed first in the jaws. Usually myelomas of the jaws appear roentgenographically as multiple rounded lesions of various sizes which have little, if any, circumferential osteosclerotic bone reaction. These vacuolated configurations are frequently situated within a larger region of diffuse radiolucency with hazy ill-defined margins. Histopathologic examination of tissue from the jaws in 5 cases disclosed tumor tissue that was composed of closely packed anaplastic cells resembling plasma cells. The supporting stroma was scanty but vascularity was prominent. This study demonstrates the prevalence of myelomas in the jaws and illustrates the contribution that oral evaluation can provide in the diagnosis of generalized disease.

2. The Effects of Varied Dosages of X-ray Irradiation on the Forming Dentition of the Ten-Day-Old Syrian Hamster, *William V. Kaplan, D.D.S., Mayo Clinic, Minneapolis, Minn.* This study was undertaken to determine by means of the roentgenogram the direct effect of varied doses of x-ray irradiation on the developing dentition of the ten-day-old Syrian hamster. In this study only the molar teeth were studied. Thirty Syrian hamsters, ten days of age were divided into groups of six animals each and the five groups were exposed to 250 h, 500 r, 1,000 r, 2,000 r, and 4,000 r respectively. Each animal was irradiated over the region of the maxillary and mandibular tooth bearing areas. A therapy type x-ray machine was used to deliver the prescribed dosages.

A littermate control was used for each irradiated animal. The controls and treated animals were killed at 100 days of age and a comparison of roentgenograms of the head and jaws of the treated and untreated animals as well as a comparison of the roentgenograms of animals treated with various dosages was carried out. Effects of irradiation on the forming dental system were: incomplete formation of roots, dwarfing of teeth and anadontia. The effects of irradiation were in direct relation to the dosage given. The left side from which the irradiation was given showed slighly greater effect on the dental structures than the opposite side. Also noted as an effect of irradiation was a retardation in the growth of the craniofacial skeleton. Various measurements of the skull and jaws were made on the roentgenograms and the several experimental series were compared with each other and also with the control series. Osseous growth and development seemed to be curtailed by irradiation. The amount of retardation of growth was in direct relation to the dosage given while the greatest underdevelopment was apparent between the group which received 500 r and 1,000 r respectively. Other gross effects of irradiation appeared to be loss of hair and hair color and necrosis of the skin in the exposed parts.